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Biologics License Application Supplement

Medical Officer's Review

Rebif® (Interferon b-1a)

FOR TREATMENT OF

RELAPSING-REMITTING MULTIPLE SCLEROSIS

Submitted by Serono, Inc.

STN #: 103780 / 0

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Overview

Interferon ß-1a (Rebif®) has been under development by Serono, Inc. for the treatment of multiple sclerosis (MS). Serono, Inc. submitted a Biologics License Application (BLA) on February 27, 1998 for the approval of Rebif® for the treatment of patients with relapsing-remitting MS (RRMS). The focus of the application was a 560-subject study that was a randomized, double-blind, placebo-controlled study of 22 µg *vs.* 44 µg Rebif® *vs.* placebo administered subcutaneously (SC) three times per week for 2 years. The application also contained additional open-label safety data from other studies. Based on the review of the BLA submission conducted by CBER, it was concluded that both doses of Rebif® were demonstrated to be safe and effective and to be approvable for treatment of RRMS. However, Serono was prevented from marketing Rebif® in the United States by the Orphan Drug Exclusivity previously granted to Biogen for the marketing of their interferon β-1a, Avonex® and to Berlex/Chiron for the marketing of their interferon β-1b, Betaseron® Betaseron®'s exclusivity expired in July 2000. Rebif® was, however, successfully approved and marketed in other countries, and thus has been commercially available in other portions of the world for several years. The subject of the current BLA amendment is a comparative study that Serono conducted to compare the efficacy of Rebif® to marketed doses of Avonex® in the treatment of relapsing-remitting MS. The study design was discussed with CBER and the design was agreed upon prior to initiation of the study. The study was a randomized, open-label study in which subjects with RRMS were treated with either Rebif® 44 µg SC three times per week or Avonex® 30 µg IM once weekly, with neurologic examinations performed by physicians blinded to the treatment assignment and with subjects' MRIs read at a central reading facility by neuroradiologists blinded to treatment assignment. Although the duration of the study was to be 48 weeks, the pre-specified primary outcome measure was the proportion of subjects who remained relapse-free following 24 weeks of treatment. Supportive evidence (secondary endpoints) were comparisons of MRI abnormalities. The Applicant conducted and submitted this BLA amendment in the belief that demonstration of superior clinical benefit of Rebif® over Avonex® would allow them to break Biogen's orphan drug exclusivity and thus, to market Rebif® in the U.S. for the treatment of relapsing-remitting MS.

Scope of this review

The focus of this document is upon efficacy information obtained primarily from a single study, XXXXXXXXXX, a randomized, open-label comparative study of the use of Rebif® 44 µg administered SC 3x per week *vs.* Avonex® 30 µg administered IM once weekly. Efficacy and safety data from the BLA previously submitted by the Applicant and reviewed by CBER for the treatment of relapsing-remitting MS will be summarized. Safety data from several additional studies of Rebif® in MS, including safety data from the Applicant's post-marketing safety database will also be summarized.

Abbreviations and Definitions of Terms Used in This Review

2-5 OAS	2', 5'-oligoadenylate synthetase
ADL	Activities of Daily Living
AE	Adverse Event
ALT	Alanine Transaminase
ANOVA	Analysis of Variance
ANCOVA	Analysis of Covariance
AST	Aspartate Transaminase
Avonex®	Biogen's recombinant human interferon β-1a
BBB	Blood-brain barrier
Betaseron®	Berlex's human interferon β-1b
CHO	Chinese Hamster Ovary
CRA	Clinical Research Associate
CRF	Case Report Form
CU	Combined Unique (T1 + T2)
DER	Drug Event Report (form)
EC	Ethics Committee
ECG	Electrocardiogram
EDSS	(Kurtkze's) Expanded Disability Status Scale
Gd	Gadolinium
Gd-DPTA	Gadolinium-diethylenetriaminepentacetic acid
HCG	Human Chorionic Gonadotrophin
HLA	Human Lymphocyte Antigen
HTLV-1	Human T-cell lymphotrophic virus, type 1
IEC	Independent Ethics Committee
IFN	Interferon
IFN β-1a	Recombinant human interferon β-1a
IM	Intramuscular (ly)
IRB	Institutional Review Board
IU	International Unit(s)
IUD	Intrauterine device
IV	Intravenous (ly)
KFS	Kurtzke Functional Systems
mcg	microgram
mg	milligram(s)
mL	milliliter
MIU	Million International Units (10^6 IU)
MR	Magnetic Resonance
MRI	Magnetic Resonance Imaging
MS	Multiple Sclerosis
MSCRG	Multiple Sclerosis Collaborative Research Group
NSAID	Non-steroidal anti-inflammatory drug

PRISMS	Prevention of Relapses and disability by Interferon β-1a Subcutaneously in Multiple Sclerosis
prn	as needed
qod	every alternate day
qw	once weekly
Rebif®	Serono's recombinant human interferon β-1a
RRMS	Relapsing-remitting multiple sclerosis
SAE	Serious Adverse Event
SC	Subcutaneous (ly)
SGOT (AST)	Serum glutamic oxaloacetic transaminase
SGPT (ALT)	Serum glutamic pyruvic transaminase
SNRS	Scripps Neurological Rating Scale
SPECTRIMS	Secondary Progressive Efficacy Clinical Trial of Recombinant Interferon-beta in Multiple Sclerosis
SPMS	Secondary progressive multiple sclerosis
T1	T1-weighted MRI scanning sequence
T2	T2-weighted MRI scanning sequence
tiw	three times per week
$t_{1/2}$	half-life
µg	microgram(s)
ULN	Upper Limit of Normal
USAN	United States Adopted Names
USP	United States Pharmacopoeia
WHO	World Health Organization
WHOART	World Health Organization Adverse Reaction Terminology

Introduction

Multiple Sclerosis

Background and Diagnostic Criteria

Multiple Sclerosis (MS) is a chronic, inflammatory, demyelinating disease of the central nervous system of unknown etiology, although an autoimmune process has often been implicated. It is a common cause of neurological disability in young adults, primarily affecting people between 20 and 40 years of age, and affects women approximately twice as often as men. Although there are sometimes inconsistencies of terminology with regard to categorization of MS within the MS field, and although some persons with MS do not neatly fit into one of the following categories, it has often proved useful to discuss MS according to clinical course, in order to: 1) provide advice on prognosis, 2) incorporate this information into consideration of potential therapeutic choices, and 3) to improve homogeneity and thus chances for detecting clinically meaningful effects in designs of clinical trials of new therapies. Most experts in the field generally recognize three clinical forms of MS: relapsing-remitting, secondary progressive and primary progressive (Lublin and Reingold, 1996). Relapsing-remitting MS (RRMS) comprises the most common form at onset. It is the presenting form in up to an estimated 80-85% of subjects, and involves recurrent attacks of neurological symptoms and signs (relapses or exacerbations) involving multiple areas of the nervous system that occur at variable time intervals ranging from months to years between attacks. These exacerbations or relapses are followed by subsequent variable degrees of recovery (remissions). The majority of subjects with RRMS develop secondary progressive MS (SPMS) in which periods of stable recovery give way to neurological decline over time. About 50% of subjects with RRMS will develop the secondary progressive form of the disease within 10 years of onset; the proportion approaches 80% after 25 years (Runmarker and Anderson, 1993). Primary progressive MS is distinct from SPMS and is characterized by steady accumulation of neurological deficit from onset, without superimposed attacks or exacerbations; it affects a much smaller percentage of subjects, perhaps 10-20% of subjects with MS present with this form.

Subjects with secondary progressive MS may continue to experience relapses; such subjects are then sometimes referred to as suffering relapsing-progressive disease; however, others (Weinshenker et al., 1989), use the term relapsing progressive MS to define subjects with a category they regard as exhibiting a mixture of primary progressive and secondary progressive disease, not simply a subset of secondary progressive MS. Such disparities in the use of terminology such as relapsing progressive account for some of the confusion in the literature regarding categorizations of MS beyond the three widely accepted categories of relapsing-remitting, secondary progressive and primary progressive disease. All experts agree, however, that there is a general tendency for the frequency of relapses to decline with time as the slowly progressive nature of the disease becomes more prominent. Weinshenker et al. in 1989 were amongst the first to provide comprehensive natural history data that supported the belief that progression of disability following conversion to SPMS is more rapid than prior to conversion.

Several phases of pathological change occur in MS, including breakdown of the blood-brain barrier with edema, lymphocytic infiltration with cytokine release, demyelination and axonal loss. Functional impairment can occur with any of these pathological processes, but irreversible deficits are thought to result from demyelination and axonal injury. There is evidence from magnetic resonance spectroscopy (Arnold et al., 1994 and DeStefano et al., 1998) and pathological studies (Trapp et al., 1998 and Ferguson et al., 1997) that axonal dysfunction and loss can occur even at early clinical stages of the disease.

Current Treatment of MS

There are currently four drugs approved for the treatment of MS in the United States. Betseron® (Interferon β-1b) and Avonex® (Interferon β-1a) have been demonstrated to have beneficial effects on relapsing-remitting MS. Betaseron® administered at a dose of 8 MIU SC every other day was demonstrated to decrease the frequency of clinical exacerbations in ambulatory patients with relapsing-remitting MS. Avonex® administered at a dose of 30 µg IM once weekly was demonstrated to decrease the frequency of clinical exacerbations and to slow the accumulation of physical disability as measured by Kurtzke's Expanded Disability Status Scale (EDSS) score. Copaxone® (glatiramer acetate – formerly known as copolymer-1) administered at a dose of 20 µg SC once daily was demonstrated to decrease the frequency of relapses in patients with relapsing-remitting MS. All three drugs have consistently been shown to reduce the frequency of clinical exacerbations compared to placebo by approximately one-third. Their effects have never previously been compared directly in a clinical trial. Novantrone® (Mitoxantrone), a cancer chemotherapeutic agent, was approved in 2000 for reducing neurologic disability and/or the frequency of clinical relapses in patients with secondary (chronic) progressive, progressive relapsing, or worsening relapsing-remitting multiple sclerosis. However, its cumulative dose-limiting cardiotoxicity restricts its role in the treatment of MS.

Other immunosuppressive agents, including cyclophosphamide, azathioprine and low-dose methotrexate have been demonstrated to have only modest effects in treating MS, and are not widely used in this country. Similarly, intravenous immunoglobulin infusions (IVIG) are felt by some to be effective in treating MS, but are not widely used in the U.S., and do not have an approved indication for the treatment of MS.

The Orphan Drug Act and Orphan Drug Exclusivity Regulations

The Orphan Drug Regulations in 21 CFR Part 316 were written to implement section 2 of the Orphan Drug Act of 1983, that consists of four sections added to the Federal Food, Drug, and Cosmetic Act. The Orphan Drug Act directs FDA to provide written recommendations on studies required for approval of a marketing application for an orphan drug. It also provides for the designation of drugs, including antibiotics and biological products, as orphan drugs when certain conditions are met, and it provides conditions under which a sponsor of an approved orphan drug enjoys exclusive approval for that drug for the orphan indication for 7 years following the date of the drug's marketing approval.

The orphan drug regulations state that "after approval of a sponsor's marketing application…FDA will not approve another sponsor's marketing application for the same drug before the expiration of 7 years from the date of such approval" (21CFR316.31(a)). The definition of "exclusive approval" clarifies that this exclusivity applies only to the same indication as the originator product's approval (21CFR316.3(b)(12)).

The definition of "same drug" (21CFR316.3(b)(13)) describes how to judge two products from different sponsors on a physical-chemical basis for different classes of molecules. For each description, however, a showing that the follow-on product is clinically superior to the originator product can supercede the structural determination of "same drug".

The definition of "clinically superior" (21CFR316.3(b)(3)) describes the criteria for judging the subsequent product clinically superior to the already approved product. A new product can be deemed clinically superior if it has shown greater effectiveness on a clinically meaningful endpoint. For this demonstration, "in most cases, direct comparative clinical trials would be necessary." Alternatively, showing greater safety is adequate to deem a product clinically superior, with only "in some cases, direct comparative clinical trials" needed for this demonstration. Additionally, "in unusual cases, a demonstration that the drug otherwise makes a major contribution to patient care" can be adequate to deem the second product clinically superior to the already approved product.

In 1993 Berlex Laboratories, in conjunction with Chiron, obtained marketing approval for their interferon β-1b product, Betaseron®, for treatment of relapsing-remitting MS. Berlex had previously obtained orphan drug designation for Betaseron® for treatment of MS, so that a 7-year period of marketing exclusivity for Betaseron® began with its approval. The clinical efficacy claim of the indication was for a decrease in the frequency of clinical exacerbations in ambulatory patients with relapsing-remitting MS.

In 1996 Biogen, Inc. obtained marketing approval for their interferon β-1a product, Avonex® for treatment of relapsing forms of MS. Biogen had previously obtained orphan drug designation for Avonex® for treatment of MS. During the review of Biogen's PLA for Avonex®, CBER determined that under the orphan drug regulations Betaseron® and Avonex® were deemed the same drug for purposes of determining marketing exclusivity when physical structure alone was considered. However, the two products were deemed different when the clinical data were considered. Avonex® treatment was found to have a significantly different and superior safety profile with regard to the incidence of injection-site skin necrosis, and was thus concluded to be a different drug from Betaseron® for orphan drug marketing exclusivity purposes. Consequently, Avonex® received marketing approval in 1996 and its own 7-year period of marketing exclusivity from its approval date. The clinical efficacy claim of Avonex®'s indication was to slow the accumulation of physical disability and decrease the frequency of clinical exacerbations in patients with relapsing forms of MS.

Like Berlex and Biogen, Serono, Inc. was granted orphan drug designation for Rebif® for the treatment of MS. When they submitted a BLA in January of 1998 for the approval of their interferon β-1a product, Rebif®, for the treatment of relapsing-remitting and "transitional"

MS, CBER determined that by structural comparison only, Rebif® was deemed to be the same product as either Betaseron® or Avonex® for orphan drug purposes, since two protein products that differ only in minor amino acid sequences or in glycosylation of the product are explicitly stated in the orphan drug regulations to be regarded as the same product. Consequently, clinical data remained the only mechanism by which Rebif® could possibly be determined to be not the "same drug" for orphan drug marketing exclusivity purposes. Following their review of the data submitted by Serono in the BLA filed in 1998, CBER concluded that Rebif® should not be given marketing approval while the existing marketing exclusivity accorded to Betaseron® (expired in 2000) and Avonex® (expires in 2003) remains in effect based on the following conclusions:

FDA determined that on a solely structural basis, Rebif®, Betaseron® and Avonex® would be deemed the same drug, making clinically based information the only avenue for distinguishing the drugs for use within the currently approved patient population. Consequently, there was no basis for determining that Rebif® is not the same drug as either Avonex® or Betaseron®, since

- Serono had not submitted any information addressing a clinical comparison of Rebif® to Betaseron®;
- Serono had not submitted adequate evidence to demonstrate that Rebif® has superior clinical efficacy or superior safety over Avonex®;
- Serono had not submitted adequate evidence to demonstrate that Rebif® makes a major contribution to patient care over Avonex®;
- Serono had not submitted adequate evidence to demonstrate that a medically plausible subset of subjects with MS, distinct from those for whom Avonex® is indicated, has been studied and shown to benefit from Rebif® where no benefit may be expected from Avonex®.

Although MS is no longer considered an orphan disease today because the prevalence is now known to be more than 250,000 in the U.S., the Orphan Drug Exclusivity regulations regarding marketing exclusivity still apply to the beta-interferons, since they were granted orphan drug status for the treatment of MS at a time when MS was estimated to affect fewer than 200,000 persons in the U.S.

Overview of Prior Clinical Studies of Rebif®

Serono has completed 6 controlled trials in MS along with a number of uncontrolled trials. Two studies were previously reviewed here in CBER in 1998-1999 as part of Serono's initial BLA application for the approval of Rebif® for treatment of relapsing-remitting MS. Study GF 6789 was a 560-subject, multicenter, randomized, placebo-controlled trial that compared two doses of Rebif® (22 µg or 44 µg) vs. placebo administered SC three times per week for 2 years. The primary endpoint was the number of protocol-defined exacerbations per subject using the following definition of an exacerbation: "the appearance of a new symptom or worsening of an old symptom, attributable to MS, accompanied by appropriate new neurological abnormality, or focal neurological dysfunction lasting for at least 24 hours in the absence of fever, and preceded by stability or improvement for at least 30 days." Rebif®

treatment significantly reduced the number at one and two years of treatment and the efficacy did not differ substantially between the two doses, nor was treatment with Rebif® associated with excessive numbers of dose-limiting toxicities. Rebif® treatment also showed a positive treatment effect on many of the secondary endpoints studied (not listed in order of importance by the Applicant prospectively), including effects on moderate/severe exacerbations, time to first exacerbation, proportion of subjects exacerbation-free, time to confirmed disability and percent progressors, decrease in MRI T2 lesion volume, decrease in combined T1 and T2 MRI "activity," and reduction in hospitalizations and steroid treatment for MS. However, it was noted in the review that the incidence of "troubling" adverse events that included cytopenias and hepatic enzyme abnormalities were notably higher for the 44 µg dose than the 22 µg dose and both were higher than the rates observed in the placebo group. The percentages of these adverse events are shown in Table 1.

Table 1: Cytopenias and Liver Enzyme Elevations in Study GF 6789

WHOART preferred term	**Placebo** N=187	**IFN 22 mg TIW** N=189	**IFN 44 mg TIW** N=184
lymphopenia	11.2%	20.1%	28.8%
leukopenia	3.7%	12.7%	22.3%
granulocytopenia	3.7%	11.6%	15.2%
thrombocytopenia	1.6%	1.6%	8.2%
SGPT increased	4.3%	19.6%	27.2%
SGOT increased	3.7%	10.1%	17.4%
hepatic function abnormal	1.6%	3.7%	9.2%

It was also noted in the review that one subject had developed a severe anaphylactoid reaction that was deemed probably due to the administration of Rebif®. It occurred approximately 1 month after treatment was begun and progressed over several days to "localized symptoms of urticaria, generalized pruritus and swollen red scars at the injection site." Subsequent skin tests were said to have shown that the subject was allergic to a component in both the active drug and placebo. There was no increase in the incidence of depression or suicidal ideation or attempt in either treatment group compared to placebo. It was concluded that hematologic and hepatic toxicity was noticeably increased in a dose-dependent manner. Some of these abnormalities resulted in decisions to stop treatment. Although severe hematopoietic events occurred with greater frequency in active treated groups, there was no increase in infections. Hepatic enzyme abnormalities appeared to be isolated laboratory abnormalities. It was also noted that there were noticeable increases of important injection site adverse events with Rebif® therapy, but rare discontinuation of treatment due to these events.

A post-marketing report of "severe anaphylaxis with recombinant interferon β1a," Rebif®, was submitted as a letter to the Editor in Neurology in 1999.

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Protocol XXXXXXXXXX

Title: An Open-Label, Randomized, Multicenter, Comparative, Parallel Group Study of Rebif® 44 µg Administered Three Times per Week by Subcutaneous Injection, Compared with Avonex® 30 µg Administered Once per Week by Intramuscular Injection in the Treatment of Relapsing-Remitting Multiple Sclerosis

Period of Study Conduct: November 1999 to February 2001

Funding: Serono, Inc.

Objectives

The primary objective as stated by the Applicant in the protocol was to demonstrate that the proportion of patients with relapsing-remitting MS who were exacerbation-free would be greater with Rebif® 44 µg administered three times per week (132 µg per week) than patients treated with Avonex® 30 µg once per week for 24 weeks.

The principal secondary objective as stated in the protocol was that the MRI-determined combined unique (CU) lesion activity would be less after 24 weeks of treatment with Rebif® 44 µg three times per week than with Avonex® 30 µg once per week.

Design

This was a multicenter, open-label, randomized, comparative, parallel group study in which up to 624 interferon-naïve subjects with relapsing-remitting multiple sclerosis (RRMS) were to be equally randomized to receive either Rebif® 44 µg administered SC three times per week or Avonex® 30 µg administered IM once per week for 48 weeks. Although all enrolled subjects were to complete 48 weeks of the treatment to which they were randomized, the efficacy outcomes were to be determined after 24 weeks of treatment.

T2-weighted and T1-weighted pre- and post gadolinium enhanced MRIs were obtained within 28 ± 4 days of beginning treatment and monthly thereafter following the start of treatment until Week 24. All pre- and post-treatment MRIs were read centrally at the

XXXXXXXXXX, where the raters received only the subject's identification number, initials, date of birth and whether or not the subject was to be scanned again.

Each center was required to have two separate physicians responsible for the management of each subject: a treating physician and an evaluating physician. The treating physician was responsible for the supervision of study drug administration, for reporting and treating adverse events and monitoring safety assessments, and was also responsible for the treatment of exacerbations and for determining whether non-MS-related factors could account for neurological worsening. The evaluating physician was responsible for neurological assessments and evaluation of exacerbations, and was responsible for determining whether an apparent exacerbation met the protocol's definition. The evaluating physician was to remain unaware of treatment assignments, adverse event profiles and any changes in safety assessments throughout the trial. Both the treating physicians and subjects were instructed not to discuss these issues with the evaluating physician.

The primary study endpoint was the proportion of subjects who were exacerbation-free at 24 weeks. Subjects were instructed to inform the study center within 48 hours of the onset of an exacerbation, and at that time, the treating physician or designate would discuss the symptoms with the subject and determine whether a neurological examination was indicated. If so, the subject would be advised to come to the center for evaluation. The evaluating physician was to determine whether or not an exacerbation meeting the protocol's definition had occurred, and would evaluate its severity based on changes in the EDSS and the Kurtzke Functional Systems (KFS) score.

If the treating physician determined that corticosteroids were necessary for treatment of the exacerbation, a standard regimen of 1.0 g/day of IV methylprednisolone for three days was to be administered.

Treatment discontinuation was mandatory in case of any of the following:

1. Pregnancy
2. Loss to follow-up
3. Use of other investigational products, or
4. Use of other approved disease-modifying therapy for MS

Treatment could be discontinued for other reasons, but subjects were asked to continue the study assessments if they discontinued from the study prematurely.

Material Source

Rebif® was supplied as a sterile solution in pre-filled syringes for subcutaneous administration. Each syringe contained 0.5 mL of solution, which consisted of 44 µg (12 MIU) of interferon β-1a, 4 mg albumin (human, USP), 27.3 mg mannitol (USP), water for injection (USP) and XXXXXXXXXX for pH adjustment. The batch numbers used in this study were: XXXXXXXXXX.

Commercially available Avonex® 30 µg for IM administration was reconstituted and administered according to the directions in the package insert. The batches used were: XXXXXXXXXX.

Randomization

Treatment assignments were determined using a computer-generated randomization list generated by the Serono Biometrics Department and were allocated through a centralized telephone randomization system. Randomization was stratified by center, with an initial block size of six followed by block sizes of four. This was done, according to the Applicant, after they performed simulations that determined the randomization block size that best prevented the potential detection of the treatment assignment for the next subject to be enrolled within a center. Randomization was to have occurred within 24 hours of a subject completing screening procedures and having been found eligible for the study.

Inclusion Criteria

Subjects were deemed eligible for participation in the study if they met the following criteria:

- Age between 18 and 55 years
- Clinically definite or laboratory-supported diagnosis of relapsing-remitting MS, according to Poser's criteria
- Two or more relapses within the preceding 24 months
- Clinical stability or improving neurological state during the four weeks prior to Study Day 1
- EDSS score of 0 to 5.5, inclusive
- Two or more lesions consistent with MS on a screening T2-weighted MRI performed within 28 ± 4 days of Study Day 1
- Adequate contraception for females of childbearing potential

Exclusion Criteria

Subjects were to be excluded if any of the following were present:

- Secondary progressive, primary progressive or progressive relapsing MS
- Prior use of interferon
- Treatment with oral or systemic corticosteroids or ACTH within 4 weeks of Study Day 1 or 7 days of the screening MRI
- Significant leukopenia, defined as a white blood cell count of <0.5 x the lower limit of normal within 7 days of Study Day 1
- Elevated liver function tests (ALT, AST, alkaline phosphatase or total bilirubin > 2 x the upper limit of normal) within 7 days of Study Day 1
- Prior cytokine or anti-cytokine therapy or glatiramer acetate within the 3 months prior to Study Day 1
- Immunomodulatory or immunosuppressive therapy within the 12 months before Study Day 1, including, but not limited to, cyclophosphamide, cyclosporin, methotrexate, azathioprine, linomide and Mitoxantrone
- Previous use of cladribine or total lymphoid irradiation

- Allergy to human serum albumin, mannitol or Gd-DTPA
- IV immunoglobulin or any other investigational drug or procedure in the 6 months before Study Day 1
- Systemic disease that could interfere with subject safety or evaluation of their MS

Treatment

Dose and Administration

Subjects enrolled in this study were to receive one of two treatments for a period of at least 48 weeks:

- Rebif® 44 µg, administered SC three times weekly, or
- Avonex® 30 µg, administered IM once weekly

Rebif® was supplied as a sterile solution in pre-filled syringes. Subjects were advised to dose in the evening to minimize the impact of any adverse event and were instructed to dose each Monday, Wednesday and Friday. Instructions for self-administration, including instruction in proper injection techniques, advice regarding rotation of injection sites and advice about avoiding injection into inflamed areas was provided prior to the start of Rebif® treatment.

Avonex® was supplied in its commercially available form: a lyophilized powder to be reconstituted with sterile water for injection. Subjects were advised to administer Avonex® by following the instructions in the package insert. Subjects were not advised as to day of the week or time of day to administer Avonex® in the protocol, but individual investigators were able to advise subjects of a recommended dosing schedule.

In Amendment #4 to the protocol dated November 9, 2000, subjects were given the option of using an auto injector to self-administer their Rebif® after completion of the 24 Week Visit.

Dose Titration

In order to minimize potential side effects at the beginning of treatment with Rebif®, a dose titration schedule was instituted. The dose administered was gradually increased over the first four weeks of treatment, with 8.8 µg 3 times per week (20% of total) for the first two weeks, 22 µg 3 times per week (50% of total) for the third and fourth weeks, and the full dose of 44 µg 3 times per week was given thereafter.

The Avonex® dose was not titrated, and was administered at 30 µg once per week beginning at Study Day 1 and continued throughout the study to subjects randomized to that treatment arm.

Dose Modification for Toxicity

Toxicity was to be graded by the treating physician according to the Modified WHO Recommendations for the Grading of Acute and Subacute Toxicities. Dose modification was not to occur in case of neurological events.

In case of Grade 1 or 2 toxicity, subjects were to be treated as deemed appropriate by the treating physician. If Grade 2 toxicity persisted, the dose of Rebif® could be reduced to 50%, but the full dose was to be resumed if at all possible.

In case of Grade 3 toxicity attributable to Rebif®, the dose could be reduced to 50% or 20% as the treating physician deemed appropriate, or interrupted until the toxicity resolved to Grade 0 or 1. An attempt to resume the full dose was to be made within 4 weeks by escalating to the next dose level (50% or 100%). If Grade 3 toxicity recurred, the dose could again be reduced or interrupted until resolution to Grade 0 or 1 and then resumed or maintained at 50% dose for the remainder of the study. Further recurrence of Grade 3 toxicity or persistence after 4 weeks' interruption would result in permanent discontinuation of treatment.

In case of Grade 4 toxicity attributable to Rebif®, subjects were to be withdrawn from treatment.

For subjects randomized to the Avonex® treatment arm, investigators were told to follow the instructions supplied in the package insert for Avonex® to make adjustments for toxicity. Investigators could also consider the use of recommendations for Rebif® dose modifications if needed.

Concomitant Therapy

Treatment of Relapses During the Study

If the treating physician determined that corticosteroids were necessary for treatment of a clinical exacerbation of the subject's MS, a standard regimen of 1.0 g/day of IV methylprednisolone for three days was to be administered. If the subject were due to have a study-related MRI scan, the scan would be performed either before beginning methylprednisolone or at least 7 days after the last dose.

Treatment of Flu-Like Symptoms

Acetaminophen (paracetamol) could be given prophylactically or to treat constitutional symptoms associated with the study drugs, such as fever, myalgia or influenza-like symptoms. Non-steroidal anti-inflammatory drugs could be given if acetaminophen did not relieve these symptoms or if a subject could not tolerate acetaminophen.

Treatment of Other Symptoms

Except for those listed in the study Exclusion Criteria, any medication that was considered necessary for a subject's welfare and that would not interfere with study treatment could be given at the treating physician's discretion and recorded in the Case Report Forms.

Treatment Compliance

Subjects were provided with diary cards on which they were to record the volume of each dose, the time of administration, adverse events, and any concomitant medications. These cards were collected and reviewed by the investigator at each study visit. The subjects also used drug accountability records to document the dispensing of study medications and the return of unused drugs and empty containers.

Evaluations Performed During the Study

The study flowchart, based on the September 30, 1999 version of the protocol is shown in Table 2.

Non-laboratory screening procedures were to have been performed within 28 ± days before the planned start of treatment, after informed consent had been obtained. Laboratory screening procedures were to have been performed within 7 days before the planned start of treatment.

On Study Day 1, the first day of study treatment, a complete physical and neurological examination, including evaluation of the subject's EDSS score, KFS score, ambulation distance and timed ambulation were to have been performed within 24 hours of the planned treatment initiation. The evaluating physician was not to refer to the subject's examination results before performing the examination. Proton density T2-weighted, pre-Gd T1-weighted and post-Gd T1-weighted MRI scans were also to be obtained at this visit; these scans were to have been separated from the screening MRIs by at least 24 hours, but not more than 32 days, and could be performed up to 3 days prior to the initiation of treatment.

Table 2: Study Procedures

Procedure	28 Days Prior to Day 1	7 Days Prior to Day 1	Study Day 1	Week 4	Week 8	Week 12	Week 16	Week 20	Week 24	Every 12 Weeks
Medical History, Physical Examination*	X		X			X			X	X
Minor Office Visit				X	X		X	X		
Neurological Examination**	X		X			X			X	X
MRI	X		X	X	X	X	X	X	X	X****

Labs* **		X		X		X			X	X
Thyroid Function Tests		X							X	X****
Antibodies to IFN-b		X							X	X****
Document Exacerbations	X	X	X	X	X	X	X	X	X	
Adverse Events			X	X	X	X	X	X	X	
Concomitant Medications	X	X	X	X	X	X	X	X	X	

*after screening, includes only weight and vital signs
** includes the EDSS, KFS, Distance Walked and Timed Ambulation Index
*** includes hematology, blood chemistries, urinalysis
****MRIs, thyroid function tests and antibodies to IFN-β to be done at Weeks 48, 72, etc. in extension study

Neurological Examinations

Measures were undertaken to try to keep the examining physician, who was to perform all neurological examinations, blinded to treatment assignment. Subjects were instructed not to disclose their treatment or anything related to their treatment regimen, to cover injection sites before neurological examinations, and not to discuss symptoms that might be related in any way to their study treatment, such as injection site reactions or influenza-like symptoms, with the evaluating physician. Evaluating physicians were instructed to communicate with subjects only about neurological matters, and to remind subjects not to discuss other matters related to treatment. Neurological examinations included evaluations of the EDSS, KFS scores, ambulation up to 500 meters and timed ambulation (evaluation of subjects walking a measured distance of 8 meters or 25 feet along a straight course as fast as they were able – the time taken was recorded). All neurological examinations were to be performed without consulting a subject's previous neurological examination.

When neurological examinations and MRI scans were scheduled for the same visit, they were to be performed on the same day whenever possible; otherwise a time difference of no more than ± 48 hours between the evaluations was permissible.

Evaluation and Treatment of Exacerbations During the Study

An exacerbation was defined as the appearance of a new symptom or worsening of an old symptom attributable to MS, accompanied by an appropriate new neurological abnormality or focal neurological dysfunction lasting at least 24 hours in the absence of fever, and preceded by stability or improvement for at least 30 days. An "appropriate new neurological abnormality" was defined as one that was consistent with the new neurological symptoms reported by the subject, whether or not accompanied by new findings in other systems. "Focal neurological dysfunction" was defined as a symptom of CNS disturbance, possibly accompanied by objective neurological findings (such as a change in sensory perception with myelitis or change in visual acuity/color perception with optic neuritis), which was felt to be consistent with an MS exacerbation. Subjects were instructed to inform the study center

within 48 hours of the onset of an exacerbation, and at that time, the treating physician or designate would discuss the symptoms with the subject and determine whether a neurological examination was indicated. If so, the subject would be advised to come to the center for evaluation. The evaluating physician was to determine whether or not an exacerbation meeting the protocol's definition had occurred, and would evaluate its severity based on changes in the EDSS and the Kurtzke Functional Systems (KFS) score, as follows:

- Mild: EDSS change of 0 to 0.5 point, with a new neurological finding and/or and increase in KFS score of one point in one to three systems
- Moderate: EDSS change of 1.0 to 2.0 points and/or an increase in KFS score of one point in four or more systems or of two points in one to three systems
- Severe: EDSS and/or KFS score increases that exceed those described for a moderate exacerbation

The evaluating physician was not to refer to the subject's previous examination(s) before performing the neurological examination for determination of an exacerbation, but if necessary, was allowed to review the subject's previous results in order to determine if the criteria for an exacerbation had been met, and if so, the exacerbation severity. If the EDSS and KFS score changes were disparate, the exacerbation was to be graded according to the greater severity.

The treating physician would determine appropriate management of the exacerbation, including whether to prescribe corticosteroid treatment, and would perform a separate assessment of exacerbation severity based on effects on the activities of daily living (ADL), using the following scale:

- Mild: little or no effect on the ADLs
- Moderate: significant impact on the ADLs
- Severe: need for hospitalization for treatment or management of the exacerbation

The treating physician or designate would conduct weekly phone checks to determine when the exacerbation reached maximum severity and began to improve. If necessary, the subject would undergo further neurological examinations; exacerbation severity would be graded according to the worst EDSS and KFS scores recorded during the exacerbation.

Subject Contacts Between Scheduled Visits

These contacts were to have occurred by telephone at Weeks 2, 6, 10, 14, 18 and 22 to determine whether any symptoms consistent with an exacerbation had occurred and whether or not an unscheduled neurological examination by the evaluating physician was needed.

Unscheduled Visits

Subjects could be seen at any time during the study for evaluation of a possible MS exacerbation or for evaluation of possible adverse events.

Procedures for and Evaluation of MRI Scans

MRIs were performed in accordance with a scanning protocol that was designed to standardize procedures across all centers. The scanning protocol specified positioning techniques, the expected range of scanning parameters, and the exact content and format required on the film and computer tapes to be sent for central analysis at the MS/MRI

Analysis Group at the XXXXXXXXXX. The MS/MRI Analysis Group had no clinical knowledge of any of the subjects whose scans they analyzed. Scan data were read, converted, and stored in the XXXXXXXXXX MS/MRI Analysis Group's computer for quantitation and analysis. All films and tapes were checked for completeness, consistency, and compliance with the MRI scanning protocol. Scans that did not meet the standards set in the protocol were rejected for analysis. Throughout the study, communication was maintained between the MRI scanning sites and the MS/MRI Analysis Group, with frequent feedback to the sites concerning the quality and consistency of scans.

Assessment of T2 Activity
A T2 active lesion was defined as any new, recurrent, newly enlarging or persistently enlarging T2 lesion; a T2 active scan was defined as a scan showing any T2 active lesions.

- New T2 lesions were those that had not appeared in any previous scan.
- Recurrent T2 lesions were those appearing at a site where earlier lesions had disappeared.
- Enlarging T2 lesions were those showing identifiable increase in size following previous stability; they cold be either newly enlarging, if enlargement was not observed on the previous scan, or persistently enlarging, if enlargement had also been noted on the previous scan

Assessment of T1 Activity
A T1 active lesion was defined as any newly enhancing, recurrent enhancing or persistently enhancing T1 lesion; a T1 active scan was defined as a scan showing any T1 active lesions.

- Newly enhancing T1 lesions were those that had not enhanced in any previous scan.
- Recurrent enhancing T1 lesions were enhancing lesions appearing at a site where earlier lesions had disappeared.
- Persistently enhancing T1 lesions were those that had enhanced on previous scans and continued to enhance on the current scan.

Assessment of Combined Unique Activity
A combined unique (CU) active lesion was defined as any lesion that was T1 active, T2 active or both; a CU active scan was defined as a scan showing any CU active lesions. Combined unique lesion counts were obtained in three steps:

- T1 active lesions were identified by sequential review of the subject's T1 scans and each was assigned a contrast identification number that was recorded in a database.
- Similarly, T2 active lesions were identified by sequential reviews of T2 scans, and each was assigned a morphological identification number.
- The two sets of scans (T1 and T2) were then reviewed together. When a T1 active lesion and a T2 active lesion were determined to be the same, the contrast and morphological identification numbers were linked to avoid double counting of simultaneous activities in single lesions. Links could involve the current, previous, or subsequent scan. Numbers of non-linked T1 active lesions, non-linked T2 active lesions and linked lesions showing both T1 and T2 activity were then combined to give counts of new CU active lesions (consisting of new CU and recurring CU lesions) and persistent CU active lesions.

Adverse Event Collection

All adverse events occurring during the study period or during the following 30 days were to have been recorded in the Case Report Form. The report was to have included the duration of the event (onset and resolution dates), severity, and estimation of its relationship to study treatment and any concomitant treatment given or other action taken. Severity was to have been assessed as "mild," "moderate," "severe" or "life-threatening" according to the WHO common toxicity criteria, and relationship to study treatment as "unlikely," "possible" or "probable" according to the WHO definitions provided in the protocol. Exacerbations and their signs and symptoms were not to have been reported as adverse events.

Determination of Antibodies to Interferon-b

Blood was to have been collected for assay to potential antibodies to interferon-β prior to administration of study drug, and at least 24 hours after the administration of study drug at 24 weeks and every 24 weeks thereafter for the duration of the study. Assays were to be performed at a central laboratory.

Efficacy Endpoints

Primary Endpoint

Proportion of subjects who were exacerbation-free after 24 weeks

Secondary Endpoints

As ranked in order of importance prospectively by the Applicant:

1. The mean number of combined unique (CU) T1 + T2 active MRI lesions per subject per scan during 24 weeks of treatment
2. The total exacerbation count per subject
3. The mean number of T2 active lesions per subject per MRI scan

Tertiary Endpoints

- Mean number of T1 active MRI lesions per subject per scan
- Proportion of CU, T2 and T1 active MRI scans per subject
- Proportion of subjects with no active CU, T2 and T1 MRI lesions during the study period

Safety Endpoints

Safety Measurements Analyzed

The following safety parameters were to be analyzed in addition to the usual detailed safety analyses:

- Incidence of development of thyroid function test abnormalities, including T3, T4 and TSH; thyroperoxidase antibody if T3, T4 or TSH was abnormal
- Incidence of development of antibodies to interferon-β

Statistical Analysis Plan

Study Day 1 was considered to be the first day when study drug was administered.

The endpoints (primary, secondary and tertiary) were all pre-specified in the protocol and in the statistical analysis plan submitted to the FDA prior to the analysis of the study results. All analyses were conducted using two-sided tests of significance, and no adjustment was made for multiplicity, as agreed with FDA in October 2000.

Determination of Sample Size

It was estimated that a sample size of 280 evaluable subjects per treatment group would provide 95% power to detect a 30% increase in the primary endpoint, the proportion of subjects exacerbation-free at 24 weeks in the Rebif® group compared to the Avonex® group. This calculation was based on a two-sided chi square test, and assumed that the type I error rate was 5% and the proportion of exacerbation-free subjects at 24 weeks would be 65% in the Rebif® group and 50% in the Avonex® group. These values were derived from the data obtained in the Prevention of Relapses and disability by Interferon-β-1a Subcutaneously in Multiple Sclerosis (PRISMS) and Once Weekly Interferon for Multiple Sclerosis (OWIMS) studies for Rebif® 44 µg three times per week. Serono used the Rebif® 44 µg once per week as an approximation of the expected Avonex® effect. The 280 subjects per treatment group would provide 99% power to detect a 46% reduction in the Rebif® group in the mean number of CU lesions per subject per scan during 24 weeks of treatment if the Applicant's assumptions were correct. This calculation was performed using a two-sided Wilcoxon rank-sum test, and assumed a Type I error rate of 5% and a common standard deviation of 0.95. The mean number of CU lesions per subject per scan during 24 weeks of treatment were assumed to be 0.42 in the Rebif® and 0.78 in the Avonex® groups. These assumptions were based on CU activity results obtained in Phase 3 Rebif® studies in similar subject populations, using 44 µg three times per week and 44 µg once per week. Assuming a 10% dropout/non-evaluable rate, 312 subjects per group or 624 total subjects were to be randomized.

Analysis Populations

Baseline and efficacy data were to be analyzed for two subject populations: the Intent-to-Treat (ITT) Population and the Evaluable Population.

The ITT Population was to include all randomized subjects for the primary efficacy parameter. Because two centers (Centers 267 and 291) that *a priori* chose not to perform MRIs on their subjects, the subjects from those two centers were excluded from the ITT efficacy population for the MRI parameters.

The Evaluable Population was to include those subjects who had no major protocol deviations and who had either completed 24 weeks of treatment or satisfied criteria specific to individual endpoints:

- For the primary endpoint (proportion of subjects exacerbation-free at 24 weeks), a subject who stopped treatment before 24 weeks would be included in the Evaluable Population if he/she had experienced an exacerbation while on treatment.

- For MRI parameters, a subject who stopped treatment before 24 weeks would be included in the Evaluable Population if he/she had had at least one post-baseline MRI scan while on treatment. Only MRI scans taken during treatment were included in the analysis of such subjects.
- For total exacerbation count at 24 weeks, all subjects who stopped treatment before 24 weeks would be included in the Evaluable Population; however, only exacerbations occurring during treatment would be included in the analysis.

The ITT Population was agreed to be the primary analysis population for all clinical and MRI outcomes in the Statistical Analysis Plan finalized November16, 2000.

Planned Interim Analysis

An interim analysis was to have been performed when half the subjects had either completed 24 weeks of treatment or withdrawn before 24 weeks. This analysis was to include assessment of the primary and main secondary efficacy measures as well as the safety data. The study could have been discontinued if interim results suggested major safety concerns or futility of continuing the study, but not because of statistically significant differences between treatment groups.

The interim analysis was eliminated by protocol Amendment 4, dated November 9, 2000, and so was never performed.

Significance Testing, Allocation of Alpha

A total overall Type I error rate of 0.05 was maintained. Since the interim analysis was not performed, no allocation of the Type I error was required for any sequential analysis procedures.

Analysis of Baseline Parameters

Baseline data were defined as the last data collected before the first injection of Rebif® or Avonex®, either on Study Day 1 or as shortly as possible before Study Day 1.

Continuous baseline parameters were to be analyzed using a two-way analysis of variance (ANOVA) model on the ranked data, with effects for treatment and center. The full analysis model using ranked data, including the main effects and treatment-by-center interaction, was to be used to test for a significant interaction. If the interaction was significant, the full model would be considered the final model. It was not expected that ANOVA model assumptions would be satisfied, but if they were, the raw data would be used in the model as the definitive analysis.

Nominal-scaled categorical baseline parameters were to be analyzed suing the Cochran-Mantel-Haenszel (CMH) general association test, and the row means score test would be used for ordinal-scaled categorical parameters. Both analyses would be adjusted for center.

If the treatment groups differed statistically in any baseline parameter, the efficacy analyses would be adjusted for this imbalance. If any baseline parameters were thought to be

clinically different between the treatment groups, the analyses of these parameters would also be adjusted for the imbalances as supportive analyses.

Primary Endpoint – Proportion of Subjects Exacerbation-Free at 24 Weeks

The primary efficacy endpoint, proportion of exacerbation-free subjects at 24 weeks, was to be analyzed using a logistic regression model. The results were to be expressed as an odds ratio adjusted for center and treatment effects using Avonex® as the comparator.

Handling of Drop-Outs or Missing Data
For subjects who withdrew from the study before Week 24 without an exacerbation (i.e., did not receive 24 weeks of treatment and were not followed up for 24 weeks), the proportion that would be considered to be exacerbation-free was estimated as follows:

- The number of subjects in each treatment group who withdrew without an exacerbation was determined.
- The proportion of exacerbation-free subjects among those with known status (i.e., had either experienced an exacerbation before Week 24 or had completed 24 weeks without an exacerbation) was determined across both treatment groups.
- The number of subjects withdrawing without an exacerbation in each treatment group who would be considered exacerbation-free was determined as the product of these two numbers (the total number of subjects in the treatment group withdrawing without an exacerbation and the overall proportion of exacerbation-free subjects). These estimates were rounded up to the next integer if the decimal part was ≥ 0.5 and rounded down otherwise.

Secondary Endpoints – MRI Analytic Methods

The main secondary efficacy endpoint was the mean number of CU active lesions per subject per scan during 24 weeks of treatment. It was to be analyzed using a nonparametric ANCOVA model with effects for treatment and center, with the baseline number of CU active lesions as the single covariate in the model.

Other secondary endpoints included the proportion of subjects with no CU active lesions, the proportion of subjects with no T2 active lesions, and the proportion of subjects with no T1active lesions. These endpoints were to be analyzed using a logistic regression model adjusted for treatment and center.

All additional MRI parameters, with the exception of the three different proportions of subjects with no active MRI lesions, were to be analyzed using a nonparametric analysis of covariance (ANCOVA) model with effects for treatment and center, with the baseline number of active lesions of the appropriate kind as the single covariate. This methodology was thought to be appropriate if parametric model assumptions were not satisfied, which was expected based on results of previous studies (PRISMS and OWIMS). If the parametric

model assumptions were confirmed, a parametric ANCOVA model would be used, with effects for treatment and center and the baseline number of active lesions as the single covariate; this model would then be considered definitive. Assuming there were no treatment-by-center interactions and that the assumption of parallelism across all combinations of treatments and centers was satisfied, the ANCOVA model with effects for treatment and center and baseline number of active lesions as a single covariate would be considered definitive. If an interaction between treatment and center was present, the interaction term would be included in the final model. If the assumption of parallelism was not satisfied, the full ANCOVA model with main effects, covariate (i.e., the baseline number of active lesions) and all interactions would be performed, eliminating all non-significant interactions; this would then be the final analysis model.

Estimation of Missing MRI Data

Centers that *a priori* chose not to perform MRIs on their subjects had those subjects excluded from all analyses of MRI endpoints.

MRI parameters for subjects who had at least one post-baseline scan but less than the complete set of six were estimated as follows (both on-treatment and off-treatment scans were included in the ITT analysis):

- The mean number of active lesions per scan was computed using all available post-baseline scans.
- The proportion of active scans was computed using all available post-baseline scans.
- If a subject's mean number of active lesions per scan was 0, then the proportion of active scans was assumed to be 0.
- The proportion of subjects with no active lesions included only subjects whose mean number of lesions per scan was 0 in the numerator. All subjects were included in the denominator for this proportion.

MRI parameters for subjects who had no post-baseline scans were estimated as follows:

- The mean number of lesions per scan was estimated to be the median of the mean number of lesions per subject per scan across both treatment groups, using data from all subjects with post-baseline MRI scans.
- If a subject's estimated mean number of active lesions per scan was estimated to be >0, the proportion of active scans was estimated to be the median of the proportions of active scans per subject across both treatment groups, using data from all subjects with post-baseline MRI scans. If a subject's estimated mean number of lesions per scan was 0, the proportion of active scans was assumed to be 0.
- The proportion of subjects with no active lesions included only subjects whose mean number of lesions per scan was 0 in the numerator. All subjects were included in the denominator for this proportion.

If a subject did not have pre-treatment CU lesion measurements (i.e., did not have an MRI performed on Study Day 1 and/or Week –4), then the missing baseline data was estimated using the overall median value at baseline for all subjects who had pre-treatment measurements.

Safety Analyses

All subjects who received at least one injection of Rebif® or Avonex® would be included in the safety analyses.

Adverse Events
Adverse event counts and numbers of subjects reporting adverse events were to be summarized for each treatment group by body system and preferred term using the WHOART 1996 Q2 dictionary. Additional adverse event summaries were to be prepared by severity and by relationship to study drug.

Premature Discontinuations
Subjects prematurely withdrawing from the study were to be listed and summarized by primary reason for withdrawal for each treatment group.

Vital Signs and Laboratory Data
Vital signs and laboratory tests at baseline and changes from baseline were to be summarized for each treatment group. Changes in laboratory tests with respect to the normal range were to be summarized for each treatment group by means of shift tables.

Development of Antibodies to Interferon-beta Over Time
Assays for neutralizing antibodies to interferon-β were to be performed and presented after all Week 48 samples were obtained.

Study Administration

A panel of five neurologists served as a liaison committee for the study. The members were XXXXXXXXXX. This group advised the company on study design and issues related to study conduct, provided advice to the study director, discussed the analysis plan and reviewed the study results.

Serono Clinical Research Associates performed study monitoring. Study set-up and coordination were the responsibility of Serono Medical Research Associates and the Therapeutic Director. Serono Medical Research Associates also handled central management of supplies. Serono's Biometrics Department performed data management and statistical analyses.

MRI analyses were performed centrally by the MS/MRI Analysis Group at the XXXXXXXXXX.

Routine clinical laboratory assessments (hematology, blood chemistries, urinalysis and thyroid function tests) were performed centrally at XXXXXXXXXX for U.S. and Canadian sites and in XXXXXXXXXX, for European sites. Assays for antibodies to interferon-β were performed centrally at XXXXXXXXXX.

Study Results

The study was conducted between November 1999 and February 2001.

Formal Protocol Modifications

The protocol dated August 13, 1999 was amended four times before February 13, 2001 (last subject, last 24 Week visit date). This application includes subject data up to the Week 24 visit, so protocol amendments approved after February 13, 2001 were not included. The amended versions of the protocol were submitted to CBER for review prior to implementation. These changes are summarized below:

- Amendment 1, dated September 30, 1999 changed entry criteria relating to previous treatments for MS following discussions with the study investigators; clarified procedures for evaluation of exacerbations and the timing of screening assessments; made several minor administrative changes to the protocol.
- Amendment 2, dated November 15, 1999 modified specifications for study drug storage to comply with regulatory requirements; corrected summary information about a previous study; made several minor administrative changes.
- Amendment 3, dated July 25, 2000 made several minor corrections to a discussion of interpretation of EDSS and made changes to the planned statistical methods.
- Amendment 4, dated November 9, 2000, permitted the use of an auto-injector for self-administration of Rebif® optional to subjects after they had completed the Week 24 visit; eliminated a planned post-Gd-enhanced MRI scan following the Week 24 visit, and eliminated the planned interim analysis.

Changes in the Conduct of the Study or Planned Analyses

The following changes were made in the planned analyses:

- In addition to the center pooling scheme described in the statistical analysis plan, for all main effect models, centers were pooled by geographic region if all subjects in a center in both treatment groups had the same response for the dependent variable. The Applicant justifies this change to the analysis plan by stating that if this pooling was not performed, it would not have been possible to assess the treatment effect for such centers as there would have been no variability within these centers. For all full effects models to assess interactions, centers were pooled by geographic region if all subjects in a center in at least one treatment group had the same response for the dependent variable. Similarly, the Applicant states that if this pooling were not performed, it would not have been possible to assess treatment by center interaction.
- For all logistic regression analyses, a full effects model (with treatment, center and treatment by center effects) was performed in order to test for interaction effects.
- The data for the number of exacerbations within 12 and 24 months from Study Day 1 was changed to within 12 and 24 months from the screening visit to be consistent with the protocol definition of eligibility at the screening visit.

- The values of MRI parameters at Study Day 1 were considered as the baseline values if both measurements at the screening visit and the Study Day 1 visit were available; otherwise, the baseline MRI parameters were considered missing observations since to determine baseline activity, both the screening and Study Day 1 MRI scans were needed. Additionally, subjects with missing baseline MRI data had their data imputed for the ITT analysis using the overall median value at baseline for all subjects who had pre-treatment measurements (i.e., had an MRI performed on both Study Day 1 and the screening visit).
- For the main secondary endpoint only using the ITT Population, to further describe the differences between treatment groups, quartiles and product limit estimates by treatment group were presented.
- For the evaluable efficacy analysis for the total exacerbation count and steroid use for exacerbation, the offset variable used in the Poisson regression model was the minimum value of the time to the major protocol deviation and time on study if the subject completed 24 weeks of the study; otherwise, if the offset variable was the minimum value of the time to protocol deviation and the time on treatment.
- The Kaplan-Meier estimates of the time to first exacerbation were presented for the 20^{th} percentile and the first quartile. In addition, the 95% confidence interval of the hazard ratio from the Cox proportional hazard model was estimated.
- Since only one hospitalization for an exacerbation occurred other than for convenience of steroid administration, no analyses were performed for the parameter of hospitalizations for exacerbation.
- The criteria of the grading scale for lymphocyte toxicity was modified to be: Grade 0 = normal range (i.e., $>0.90 \times 10^9$/L); Grade 1 = 0.90 to 0.75 x 10^9/L; Grade 2 = 0.74 to 0.50 x 10^9/L. The grading scale for the "Lymphocytes Decreased" was modified from that described in the statistical analysis plan because the lower limit of the reference range for lymphocytes for the central laboratory was 0.91 x 10^9/L. The statistical analysis plan indicated that the lower limit of the reference range was 1.0 x 10^9/L and thus needed revision to be consistent with the laboratory's lower limit.
- Sensitivity analyses were performed for the ITT Population analyses of the primary and the main secondary parameters at baseline and during the study.
- Post-hoc analyses of certain adverse events and concomitant medications were performed using Fisher's exact test in order to detect statistically significant treatment differences between groups.
- According to the Applicant, Amendment 3 provided more precise guidelines on determination of EDSS using an adjustment for visual functional scale score as used in other clinical trials of MS and to clarify ambulation distance and its use in EDSS determination.

Protocol Deviations/Violations

Violations of Eligibility Criteria

Three subjects, all randomized to Avonex® treatment, failed to meet one specific eligibility criterion: subject XXXXXXXXXX failed the MRI inclusion criteria, subject XXXXXXXXXX had received prior treatment with interferon, and subject XXXXXXXXXX received steroid treatment within 4 weeks of Study Day 1.

Violations that Occurred During the Conduct of the Study

The following 35 violations (16 in the Rebif® treatment group, 19 in the Avonex® treatment group) that occurred after subjects had been randomized:

- 2 subjects randomized to Rebif® and 1 randomized to Avonex® missed more than 25% of their prescribed study medication
- 7 subjects randomized to Rebif® and 16 subjects randomized to Avonex® received steroid treatment within 7 days prior to an MRI scan
- 2 subjects randomized to Rebif® used a prohibited medication during the study (concomitant medications for treatment of cancer, the other received steroids)
- 1 subject randomized to Rebif® and 1 subject randomized to Avonex® received corticosteroids for more than 30 consecutive days
- 4 subjects randomized to Rebif® and 1 subject randomized to Avonex® became pregnant during the study

Reviewer's Note: *Note that 6 subjects are counted twice in the above summaries. Two subjects in the Rebif® treatment group were counted both for steroid use for more than 30 consecutive days and for steroid treatment within 7 days prior to an MRI scan, and another was counted both for use of a prohibited medication and for receiving steroid treatment within 7 days prior to an MRI scan. One subject in the Avonex® group was counted both for steroid treatment within 4 weeks of Study Day 1 and for steroid treatment within 7 days prior to an MRI scan, one subject was counted both for steroid treatment within 7 days prior to an MRI and for becoming pregnant during the study, and one subject was counted both for steroid treatment within 7 days prior to an MRI and for steroid use for more than 30 consecutive days.*

In addition, the Applicant following requests from CBER identified the following deviations/violations of protocol conduct:

- 20/676 subjects (excludes the subject randomized to Avonex® who never received study drug), or 3.0% had reversal of roles between their treating and evaluating physicians, 10 randomized to each treatment group as follows: in 3 instances, the evaluating physician became the treating physician, all at Week 24, the last study visit scheduled. In 8 instances, the treating physician at Study Day 1 (in some instances also did the Study Day 1 evaluations, and thus, functioned as both the treating and evaluating physician at this visit) became the evaluating physician for the duration of the study. In the 7 instances (of the 20 in which there was reversal of roles) in which relapses occurred, there were two cases, both in the Avonex® treatment arm, in which the evaluating physician served as both the treating and evaluating physician at an unscheduled visit for evaluation and decision regarding treatment of a relapse.

Reviewer's Note: *The only two instances where reversal of physician roles may have had an impact on the study results is in the two cases in the Avonex® group where the evaluating and treating physician were the same. This very small number of instances in which unblinding of the evaluating physician occurred is unlikely to have affected the overall study results.*

Number of Relapses Pre-Study
The Applicant notes that 16 subjects did not have 2 relapses within 2 years of study entry (5 in the Rebif® group, 11 in the Avonex® group – see Table 6), and states that although the intent was for 2 relapses within 24 months of Study Day 1, that a reading of the protocol and CRF could be interpreted as within 24 months of screening. However, the Applicant further notes that 5 subjects failed even this criterion; four were granted sponsor exemptions and one had two pre-study relapses collapsed into one based on less than 30 days between onset dates of the two reported relapses. The other 11 subjects had situations similar to the following example: a subject seen on April 21, 2000 with a history of one relapse in 1999 and a second relapse in April 1998 was considered by the site to be eligible based on 24 months prior to screening. However, the study algorithm for unknown dates for relapse onset used the first day of the month, making a subject such as this fall outside the eligibility window.

The Applicant did not report any other violations or deviations from the protocol. They specifically reported that no subject received the wrong study treatment or incorrect dose.

***Reviewer's Note**: The case report form (CRF) clearly asks "How many exacerbations have occurred within the 24 months prior to Study Day 1?" making the comment made by the Applicant above not actually relevant. However, the protocol inclusion criterion does state "two or more relapses within the prior 24 months," not specifying the 24 months prior to Study Day 1 rather than prior to the Screening Visit. Thus these inconsistencies could easily have been interpreted differently by the various study sites, accounting for 16 subjects not having had relapses within 24 months of Study Day 1.*

The overall small number and types of protocol violations/deviations did not affect the study outcome. This is shown later in the analyses on the "Evaluable Population" that excludes subjects with significant protocol deviations or violations. Furthermore, the difference between two relapses within 24 months or 25 months is not likely to be a significant influence on future course. The results of analyses on both the Evaluable and ITT Populations are statistically robust.

Study Conduct at Specific Study Sites

The Applicant informed CBER that they had been informed by the Principal Investigator (PI) of some significant irregularities at his study site (Site #238), including alleged forging of signatures of the PI and sub investigators by a clinical study coordinator who is no longer employed by the study site. The Applicant conducted a clinical site audit and recommended excluding clinical data from this site. Therefore, both CBER and the Applicant performed data analyses on the primary endpoint as shown above, both including and excluding the 11 subjects enrolled at Site #238. The results remained statistically robust and were not altered in any meaningful way by excluding these subjects from the analyses.

The three study site inspections performed by CBER's Bioresearch Monitoring inspection teams (BIMO) at Site #s 225, 236 and 242 revealed no significant deficiencies of study site conduct. The deviations noted on the FDA Form 483 issued to the investigator at Site #225 were not considered to be substantive. The data submitted by the three sites audited were considered to be reliable and accurate.

Subject Enrollment and Disposition

According to the study design, 624 subjects were planned to be enrolled in the study. By late June 2000, the Applicant felt that full enrollment would likely be reached between July 7th and July 14th. In order to ensure complete enrollment, the Applicant decided to keep enrollment open until July 14th. During the final week enrollment exceeded the expected rate, resulting in a total of 677 subjects being enrolled in study XXXXXXXXXX.

Of the 677 subjects randomized, 339 received Rebif® 44 µg SC 3 x per week and 338 subjects were assigned to receive Avonex® 30 µg IM once per week. Four hundred forty-three subjects (65%) were enrolled at 37 study sites in the U.S., 73 subjects 11%) were enrolled at 4 sites in Canada, and 161 subjects (24%) were enrolled at 15 study sites in Europe.

At the 56 total study sites, 32 sites enrolled 10-20 subjects; 19 enrolled fewer than 10 subjects, with 11 enrolling 6-9, and 8 enrolling 5 or fewer. Only 5 sites enrolled more than 20 subjects; the largest number enrolled at a single site was 24, and two others enrolled 23 subjects each; one site each enrolled 21 or 22 subjects.

Adherence to Protocol-Required Contacts Between Clinic Visits

For the 331 subjects randomized to the Rebif® treatment group and the 328 subjects randomized to Avonex® who completed the 24 weeks of the study, the mean portion of phone contacts that were completed were 84.2 and 84.6% for the two treatment groups, respectively (median portion for both 100%, ranges 0-100% for both groups). For the 8 subjects randomized to Rebif® and the 9 subjects randomized to Avonex® who did not complete the 24 weeks of treatment, the portion of the expected numbers of phone contacts that were completed were 47.9 and 82.6%, respectively (median portion 42% for Rebif® and 100% for Avonex®, ranges 0-100 for Rebif® and 33-100% for Avonex®).

Balance Across Treatment Centers

Twenty-one of the 56 centers (37.5%) had equal numbers of subjects assigned to the two treatment groups. Most of the remaining centers had treatment group imbalances of 1 subject (27 sites, 48%). Of the 8 sites where the treatment groups had an imbalance of more than one subject, 7 had imbalances of 2 subjects, and one had 3 subjects randomized to Rebif® and none to Avonex®.

Randomization

There were no errors in randomization. One subject randomized to Avonex® did not receive treatment. All 339 subjects randomized to Rebif® treatment received Rebif®.

Time on Study

Three hundred twenty-two subjects (95%) randomized to Rebif® completed 24 weeks of treatment, and of the 17 subjects who prematurely withdrew, 9 (2.7%) continued in the study for 24 weeks.

Three hundred twenty-six (96.4%) of the subjects randomized to Avonex® completed 24 weeks of treatment, and of the 12 who prematurely withdrew from treatment, 2 (0.6%) continued in the study for 24 weeks.

Time on Treatment:

Ninety-five percent of the 339 subjects randomized to Rebif® completed 24 weeks of Rebif® treatment. Three hundred twenty-six (96.4%) of the 338 subjects randomized to Avonex® completed 24 weeks of Avonex® treatment. This is shown in Table 3.

Table 3: Subject Disposition

	Rebif	Avonex
N randomized	339	338
Number Who Completed 24 Weeks of Treatment	322 (95%)	326 (96.4%)
Number Who Prematurely Discontinued Treatment	17 (5%)	12 (3.6%)
Adverse Event	11 (3.2%)	3 (0.9%)
Lack of Efficacy	1 (0.3%)	0
Subject Decision	3 (0.9%)	5 (1.5%)
Pregnancy	2 (0.6%)	0
Lost to Follow-Up	0	3 (0.9%)
Other	0	1*

*withdrew after randomization, but prior to initiation of treatment due to experiencing a relapse

Study drug compliance rates as reported on the subject diary cards and by drug accountability records are shown in Table 4.

Table 4: Treatment Compliance

		Rebif	Avonex
Number of Injections	Percent Compliant	N = 339 (%)	N = 338 (%)
	80%	328 (97))	334 (99)
	90%	322 (95)	326 (96)
	99%	231 (68)	157 (46)
Volume of Injections	80%	314 (93)	326 (96)
	90%	300 (88)	298 (88)
	99%	102 (30)	115 (34)

As Table 4 shows, compliance was 90% or greater in ≥88% of subjects in both treatment groups, and in ≥ 95% of subjects in both treatment groups if judged by the number of injections recorded in the subject diaries. Relatively few subjects in both treatment groups were ≥99% compliant. It should be noted that some of the differences in compliance rates can be accounted for by the difference in treatment regimens, since a subject randomized to Avonex® who missed one of the 24 scheduled injections in 24 weeks would be counted as being 96% compliant, whereas a subject randomized to Rebif® who missed one of the 72 scheduled injections in 24 weeks would be counted as being 99% compliant.

There were 45 subjects in the Rebif® treatment group who had dose modifications during the study. Thirty-one (9.1%) subjects had their dose reduced (8 for elevation of their hepatic enzymes), 14 (4.1%) had their dose interrupted (2 for elevation of their hepatic enzymes, 2 for fatigue, 2 for influenza-like symptoms, 1 for injection site pain), and 11 (3.2%) had their dose discontinued for adverse events. Twenty-six (7.7%) subjects in the Avonex® treatment group had dose modifications: 12 (3.6%) had their dose reduced, 12 had their dose interrupted (2 for chest pain, 1 for neutropenia, 1 for convulsions), and 3 (0.9%) had their dose discontinued for adverse events.

Adverse Events Leading to Premature Discontinuation

Fourteen subjects were identified as discontinuing from the study due to 26 adverse events (2% overall). Eleven of these, and 21 of the adverse events were in the Rebif® group (3.2% of the subjects randomized to Rebif®, and 3 with 5 adverse events were in the Avonex® group (0.9%). These are shown in Table 5.

Table 5: Adverse Events Resulting in Premature Study Discontinuation

~~Treatment Group~~	~~Subject ID~~	~~Adverse Event(s)~~
Rebif 44 ug TIW	2140001	Flu-Like Symptoms, Injection Site Reactions
	2720006	Flu-Like Symptoms, Injection Site Burning, Depression
	2490002	Flu-Like Symptoms, Skin Tactile Sensitiviity, Fatigue, Decreased Balance
	2740014	Flu-Like Symptoms, Depression, Burning at Injection Site
	2560001	Injection Site Burning
	2460001	SGOT, SGPT Increase
	2800004	Transaminases Increased
	2800008	Lymphopenia, Leukopenia
	2740016	Anorexia
	2760012	Palpitations, Pain in Extremities
	2780001	Severe Pain in Legs
Avonex 30 ug QW	1980003	Depression, Insomnia
	2720010	Paraparesis
	2740003	Urticaria, Anorexia, Weight Loss

Eight subjects were classified as prematurely discontinuing from the study primarily due to "patient decision." Six of these 8 subjects also had ongoing adverse events at the time of study discontinuation, 2 in the Rebif® group, 4 in the Avonex® group. In addition, 2 of the 3 subjects classified as having prematurely discontinued from the study due to being "lost to follow-up" had ongoing adverse events at the time of study discontinuation. Both subjects were receiving Avonex®.

Demographics and Baseline Characteristics

Table 6 shows the baseline demographic characteristics of the subjects enrolled in the trial. Subjects had a mean age of 37-38 years, were predominantly white (90-92%) females (75%). The treatment groups were well balanced with regard to baseline demographics.

Table 6: Baseline Demographic Characteristics

	Rebif	Avonex
N	339	338
Age (years)		
<20	6	2
20-29	51	62
30-39	125	138
40-49	114	105
50-55	43	31
Mean (SD)	38.3 (9.0)	37.4 (8.6)
Sex		
Female	254 (74.9%)	252 (74.6%)
Male	85 (25.1%)	86 (25.4%)
Race		
White	313 (92.3%)	303 (89.6%)
Black	13 (3.8%)	23 (6.8%)
Hispanic	7 (2.1%)	7 (2.1%)
Asian	0	1 (0.3%)
Other	6 (1.8%)	4 (1.2%)
BMI		
mean (SD)	26.6 (6.1)	26.7 (5.7)
median	25.4	25.9

Table 7 shows subject's baseline characteristics with regard to duration of disease, number of exacerbations in the previous one and two years and EDSS scores at baseline. Again, the treatment groups were well balanced on these characteristics.

Table 7: Baseline Characteristics of Subjects' MS

Characteristic	Rebif	Avonex	p-value
	N = 339	N = 338	
Time Since Onset of MS			0.592*
N	339	336	
Mean(yrs)	6.5 (6.4)	6.7 (6.5)	
Median (SD)	4.0	4.1	
Range	0.4, 37.5	0.3, 34.9	
Time Since Most Recent Exacerbation (mos)			0.449*
Mean(SD)	5.2 (3.4)	5.0 (3.1)	
Median	4.4	3.9	
Range	0.1, 21.8	1.1, 18.9	
No. of Exacerbations Within 12 mos. Of Study Day 1			0.488**
0	13 (3.8%)	10 (3.0%)	
1	110 (32.4%)	125 (37.0%	
2	159 (46.9%)	157 (46.4%)	
3	49 (14.5%)	36 (10.6%)	
>=4	8 (2.4%)	10 (3.0%)	
No. of Exacerbations Within 24 mos. Of Study Day 1			0.442**
<2	5 (1.5%)	11 (3.3%)	
2	184 (54.3%)	192 (56.8%)	
3	105 (31.0%)	90 (26.6%)	
4	33 (9.7%)	30 (8.9%)	
>=5	12 (3.5%)	15 (4.4%)	
EDSS score			0.310**
0	17 (5.0%)	24 (7.1%)	
1	90 (26.5%)	67 (19.8%)	
2	117 (34.5%)	138 (40.8%)	
3	67 (19.8%)	64 (18.9%)	
4	37 (10.9%)	36 (10.7%)	
5	11 (3.2%)	9 (2.7%)	
Received Treatment for MS Within 12 mos. Of Study Day 1			
Yes	167 (49.3%)	154 (45.6%)	
No	172 (50.7%)	184 (54.4%)	

Similarly, the MRI characteristics at baseline were well balanced between the two treatment groups as shown in Table 8.

The values for the 25th percentile, median and 75th percentile were identical for both treatment groups (0.0, 1.0, 2.0) at indicating that the groups were balanced in terms of CU active lesions at baseline. The mean values were different at baseline, 2.4 (4.7) for Rebif® and 2.9 (7.1), largely because of the extreme outlier in the Avonex® group with 83 T1 active (and therefore also CU active) MRI lesions at baseline. When this one outlier from the Avonex® group was excluded, the means were 2.4 (4.7) for Rebif® and 2.6 (5.6) for Avonex®.

Table 8: Baseline MRI Characteristics

	Rebif	Avonex
	N= 325	N = 325
Number of CU Lesions per Subject		
No CU Active Lesions	146 (44.9%)	147 (45.2%)
Mean (SD)	2.4 (4.7)	2.9 (7.1)
Median	1.0	1.0
Range	0.0, 33.0	0.0, 83
Missing	16	20
Number of T2 Active Lesions per Subject		
No T2 Active Lesions	201 (61.8%)	205 (63.1%)
Mean (SD)	1.2 (2.6)	1.1 (2.5)
Median	0.0	0.0
Range	0.0, 19.0	0.0, 15.0
Missing	16	20
Number of T1 Active Lesions per Subject		
No T1 Active Lesions	186 (57.2%)	178 (54.8%)
Mean (SD)	1.9 (4.2)	2.5 (7.0)
Median	0.0	0.0
Range	0.0, 30.0	0.0, 83.0
Missing	16	20

Reviewer's Note: *A T2 active scan was defined for this study as being a scan that had at least one T2 active lesion (new, newly enlarging, and persistently enlarging T2 lesions). The baseline MRI scan was performed within 3 days of Study Day 1 and is compared to the Screening MRI performed within 28 days of Study Day 1, explaining how subjects can have "active T2 lesions" at baseline.*

Note that nearly half the subjects had no combined unique active lesions on MRI at baseline (new or enlarging lesions) compared to their Screening MRI.

Note also the extreme outlier value of 83 T1 active lesions in one subject in the Avonex® group. This outlier needs to be taken into account in the analyses performed to assess treatment effects on the secondary endpoint of mean CU MRI lesion activity.

Efficacy Results

The Intent-to Treat (ITT) Population, consisting of all subjects randomized, with one exception only, was the primary analysis population for all clinical and MRI outcomes. That exception was determined prospectively, and applied only to the MRI outcomes. Two centers, Center 267 that enrolled 5 subjects, Center 291 that enrolled 22 subjects had been granted *a prioi* permission not to perform MRI scans on their subjects, and so these subjects were excluded from the ITT efficacy population for the MRI parameters. Analyses of the Evaluable Population were used to support the ITT analyses. The Evaluable Population was defined as including all subjects who had no major protocol violations and who had either completed 24 weeks of treatment or satisfied criteria specific to individual endpoints, defined as follows:

- For the primary endpoint (proportion of subjects exacerbation-free at 24 weeks), a subject who stopped treatment before 24 weeks would be included in the Evaluable Population if he/she had experienced an exacerbation while on treatment.
- For the MRI parameters, a subject who stopped treatment before 24 weeks would be included in the Evaluable Population if he/she had had at least one post-baseline MRI scan while on treatment. Only MRI scans taken during treatment were included in the analysis for such subjects.
- For total exacerbation count at 24 weeks, all subjects who stopped treatment before 24 weeks would be included in the Evaluable Population; however, only exacerbations occurring during treatment would be included in the analysis.

Primary Endpoint Results

During the 24-week treatment period, 74.9% of subjects in the Rebif® treatment group and 63.3% of subjects in the Avonex® treatment group remained exacerbation-free.

Table 9: Overall Results on Primary Endpoint

	Rebif	Avonex
	N = 339	N = 338
	N (%)	N (%)
Exacerbation-Free	254 (74.9)	214 (63.3)
Not Exacerbation-Free	85 (25.1)	124 (36.7)
	Treatment Comparison	
Odds Ratio (O.R.)	1.9	
95% CI of O.R.	1.3, 2.6	
p-value	<0.001	
Relative Risk (RR)	1.18	
95% CI of RR	1.07, 1.31	

The Applicant performed a sensitivity analysis in which all premature discontinuations from the Rebif® group without exacerbation information was considered to have experienced a relapse and all Avonex® dropouts without exacerbation information were considered to be relapse-free. Because adjustments were very small, 4 in the Rebif® group and 8 in the Avonex® group, the between group comparison remained significant (p=0.0055; adjusted odds ratio 1.6; 95% CI of the odds ratio = 1.2, 2.3).

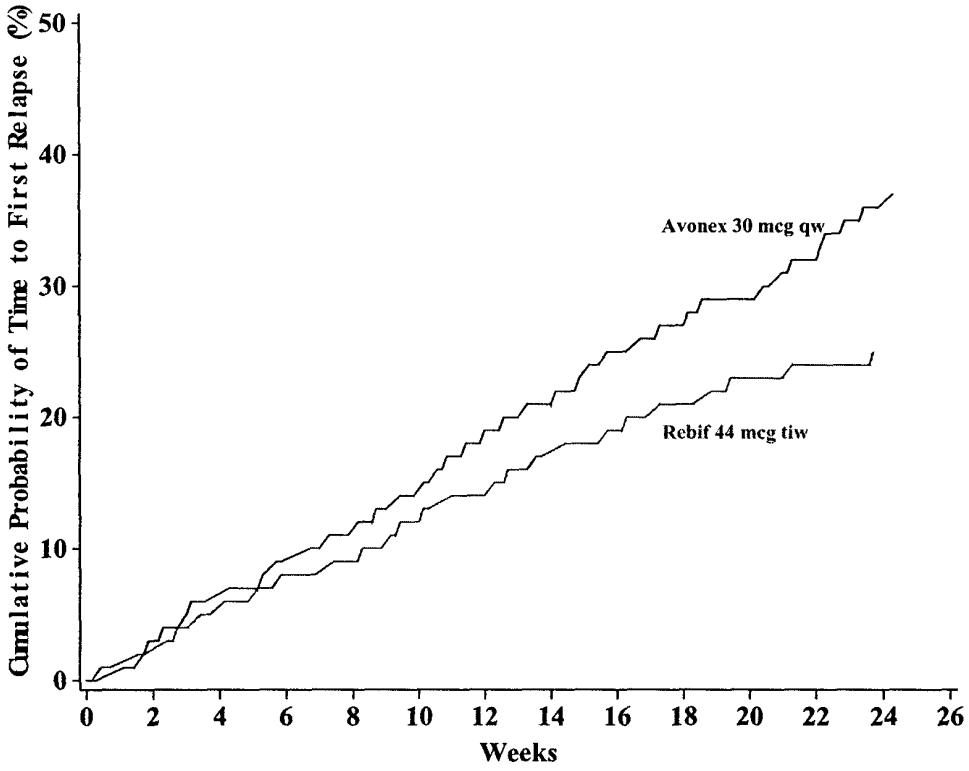
The results of the ITT analysis were confirmed by the Applicant for the Evaluable Population, and also demonstrated a significant difference in favor of Rebif® (p = 0.003 on both the RR and O.R.).

Table 10: Evaluable Population Analysis of Primary Endpoint

	Rebif	Avonex
	N = 322	N= 325
	N (%)	N (%)
Exacerbation Free	239 (74.2)	207 (63.7)
Not Exacerbaton Free	83 (25.8)	118 (36.3)
	Treatment Comparison	
Odds Ratio (O.R.)	1.7	
95% CI of O.R.	1.2, 2.3	
p-value	0.003	
Relative Risk (RR)	1.17	
95% CI of RR	1.05, 1.30	

The Applicant as shown in Figure 1 explored this further. This demonstrates the cumulative proportion of subjects experiencing a relapse over time after starting interferon therapy. It shows that there is a 32% relative reduction in the proportion of Rebif® subjects who experience relapses compared to Avonex®-treated subjects over the 24 weeks of treatment, an effect that does not appear to be lessening at 26 weeks.

Figure 1: Cumulative Proportion of Subjects to Time of Next Relapse



Unscheduled Visits and Determination of Exacerbations During the Study

Two hundred one visits occurred at which a neurological examination was performed when it was not scheduled in the protocol. Seventy-six of these were performed at an otherwise scheduled visit (at the "minor" office visits) and 125 at a completely unscheduled visit. One hundred nineteen unscheduled visits occurred at which no neurological assessments were performed. These involved visits for repeat laboratory testing, adverse event assessments, follow-up of a prior relapse, termination of treatment, injection training and for miscellaneous other reasons.

The number of subjects seen for unscheduled neurological evaluations because of concern about a possible MS exacerbation is shown in Table 11.

Table 11: Neurological Exams and Evaluations of Exacerbations

	Rebif	Avonex
Total Exacerbations	N = 98	N = 132
Scheduled Visits/Scheduled Neuro	N = 656	N = 653
# of Exacerbations (% of Exams)	28 (4.3%)	49 (7.5%)
% of Total Exacerbations	29%	37%
No Steroids (% of Exacerbations)	25 (89%)	36 (73%)
Steroids (% of Exacerbations)	3 (11%)	13 (26%)
Scheduled Vists/Unscheduled Neuro	N = 35	N =41
# of Exacerbations (% of Exams)	30 (86%)	35 (85%)
% of Total Exacerbations	31%	26%
No Steroids (% of Exacerbations)	18 (60%)	18 (51%)
Steroids (% of Exacerbations)	12 (40%)	17 (48%)
Unscheduled Visits	N =55	N =70
# of Exacerbations (% of Exams)	40 (73%)	48 (68%)
% of Total Exacerbations	41%	36%
No Steroids (% of Exacerbations)	24 (60%)	19 (40%)
Steroids (% of Exacerbations)	16 (40%)	29 (60%)

As shown in Table 11, subjects in the Avonex® group were seen more often for unscheduled visits than subjects in the Rebif® group and also had more unscheduled neurological assessments performed at scheduled visits (the "minor" office visits) during which a neurological assessment was not required by the protocol. Note that the greatest Rebif® - Avonex® difference in determination of exacerbation rates occurred at those protocol-required visits during which neurological examinations were scheduled and required, when one might expect the most rigorous ascertainment of exacerbations to have occurred.

Exacerbations determined to have occurred at both scheduled and unscheduled visits where a neurological exam had not previously been scheduled were treated with steroids in 40% of subjects in the Rebif® treatment group; in the Avonex® group exacerbations were more often treated with steroids at these types of visits regardless of whether the visit was scheduled (48.6%) or unscheduled (60.4%). At scheduled visits during which a neurological examination was also scheduled and an exacerbation was determined to be occurring, exacerbations were treated with steroids twice as often in the Avonex® group as in the Rebif® group. Note however, that the overall frequency of steroid use for treatment of these exacerbations was less in both treatment groups compared to treatment of exacerbations determined at unscheduled neurological exams. This is discussed further in the section on exploratory analyses assessing exacerbation severity.

CBER-Performed Analyses on the Primary Endpoint

Additional analyses on the primary endpoint were performed by CBER to examine for differences in response to treatment by various subgroups that included treatment by sex, age, and half of the study into which the subjects were enrolled and geographical differences (U.S. vs. Canada vs. Europe).

The numbers of male and female subjects experiencing no relapses and those who experienced at least one relapse is shown in Table 12. The table shows that Rebif® treatment increases the proportion of both female and male subjects who are exacerbation-free, and does not increase the proportion of subjects of either sex who do experience exacerbations.

Table 12: Number of Subjects Exacerbation-Free by Sex

Males:

	REBIF	AVONEX
	N = 85	N = 86
Exacerbation-free	69 (81%)	56 (65%)
Not exacerbation-free	16 (19%)	30 (35%)

Females:

	REBIF	AVONEX
	N = 254	N = 252
Exacerbation-free	185 (73%)	158 (63%)
Not exacerbation-free	69 (27%)	94 (37%)

The CMH p-value for treatment effect stratified by sex is 0.0011. The estimates for overall odds ratio and relative risk based on this stratified analysis are:

Odds Ratio: 1.7 95%CI: [1.2, 2.4]
Rel. Risk: 1.19 95%CI: [1.07, 1.31]

Excluding site #238 the p-value is 0.0003.

An analysis stratified by age, using approximately both the median and mean subject age of 38 years, revealed no important differences in response to treatment of those younger than age 38 vs. those who were older than 38 years, as shown in Table 13.

Table 13: Number of Subjects Exacerbation-Free by Age

Age < 38 years:

	REBIF	AVONEX
	N = 157	N = 180
Exacerbation-free	110 (70%)	108 (60%)
Not exacerbation-free	47 (30%)	72 (40%)

Age = 38 years:

	REBIF	AVONEX
	N = 182	N = 158
Exacerbation-free	144 (79%)	106 (67%)
Not exacerbation-free	38 (21%)	52 (33%)

The CMH p-value for treatment effect stratified by age is 0.0017. The estimates for overall odds ratio and relative risk based on this stratified analysis are:

Odds Ratio: 1.7 95%CI: [1.2, 2.4]
Rel. Risk: 1.17 95%CI: [1.06, 1.30]

Excluding site #238 the p-value is 0.0006.

Note that in this table there appears to be an age imbalance between the treatment groups that is not apparent in the demographic data presented in Table 6. This is due to the small numbers of subjects 37 and 38 years of age in the Rebif® treatment group (8 and 6, respectively) and a larger number of subjects 37 years of age (17) and smaller number of subjects 38 years of age (7) in the Avonex® treatment group.

When treatment by date of enrollment by study halves was examined, it revealed that there were no important overall differences in results, as shown in Table 14.

Table 14: Treatment by Enrollment Date

First Half of Study:

	REBIF	AVONEX
	N = 172	N = 166
Exacerbation-free	132 (77%)	103 (62%)
Not exacerbation-free	40 (23%)	63 (38%)

Second Half of Study:

	REBIF	AVONEX
	N = 167	N = 172
Exacerbation-free	122 (73%)	111 (65%)
Not exacerbation free	45 (27%)	61 (35%)

The CMH p-value for treatment effect stratified by study half enrollment is 0.0011. Excluding site #238 the p-value is 0.0004.

The numbers of subjects who were exacerbation-free or not exacerbation-free by geographical area (U.S., Canada and Europe) are shown in Table 12. There were no overall differences from the effects seen on the population as a whole. This analysis again was highly statistically significant, $p<0.001$.

Table 15: Exacerbations During 24 Months by Geographical Location of Study Site

United States:

	REBIF	AVONEX
	N = 223	N = 220
Exacerbation-free	173 (78%)	144 (65%)
Not exacerbation-free	50 (22%)	76 (35%)

Canada:

	REBIF	AVONEX
	N = 35	N = 38
Exacerbation-free	24 (69%)	24 (63%)
Not exacerbation-free	11 (31%)	14 (37%)

Europe:

	REBIF	AVONEX
	N = 81	N = 80
Exacerbation-free	57 (70%)	46 (58%)
Not exacerbation-free	24 (30%)	34 (42%)

The CMH p-value for treatment effect stratified by geographical location is 0.0011. Excluding site #238 the p-value is 0.0004.

Impact of Baseline MRI Lesion Status on the Primary Efficacy Endpoint*:*
CBER performed several ancillary stratified analyses to assess the robustness of the Applicant's findings for the primary endpoint applying a categorical adjustment for baseline CU, T1, and T2 lesion counts. In these analyses, strata for baseline lesion counts of each type were constructed using the particular overall median baseline lesion count (i.e., for both treatment groups combined). One of the reasons this approach was taken was that there were two outliers (viz., baseline T1 lesion counts of 42 and 83) which occurred in two Avonex®-treated subjects. Analyses were also performed excluding the problematic center, #238.

Baseline CU Lesion Count £ 1:

	REBIF	AVONEX
	N = 219	N = 217
Exacerbation-free	176 (80%)	140 (65%)
Not exacerbation-free	43 (20%)	77 (35%)

Baseline CU Lesion Count > 1:

	REBIF	AVONEX
	N = 106	N = 108
Exacerbation-free	68 (64%)	66 (61%)
Not exacerbation-free	38 (36%)	42 (39%)

The two-sided Cochran Mantel-Haenszel test yields a p-value of 0.0013. Excluding site #238 yields a p-value of 0.0004.

Baseline T1 Lesion Count £ 0:

	REBIF	AVONEX
	N = 186	N = 178
Exacerbation-free	149 (80%)	114 (64%)
Not exacerbation-free	37 (20%)	64 (36%)

Baseline T1 Lesion Count > 0:

	REBIF	AVONEX
	N = 139	N = 147
Exacerbation-free	95 (68%)	92 (63%)
Not exacerbation-free	44 (32%)	55 (37%)

The two-sided Cochran Mantel-Haenszel test yields a p-value of 0.0014. Excluding site #238 yields a p-value of 0.0005.

Baseline T2 Lesion Count £ 0:

	REBIF	AVONEX
	N = 201	N = 205
Exacerbation-free	157 (78%)	136 (66%)
Not exacerbation-free	44 (22%)	69 (34%)

Baseline T2 Lesion Count > 0:

	REBIF	AVONEX
	N = 124	N = 120
Exacerbation-free	87 (70%)	70 (58%)
Not exacerbation-free	37 (30%)	50 (42%)

The two-sided Cochran Mantel-Haenszel test yields a p-value of 0.0011. Excluding site #238 yields a p-value of 0.0003.

In summary, CBER confirmed the analyses performed by the Applicant on the primary study endpoint, and all subset analyses are consistent with the overall study results.

Secondary Endpoints

MRI-Determined CU Activity

The main secondary objective of the study was to demonstrate that MRI-determined combined unique (CU) activity (that is, any MRI lesion that was T1 active, T2 active or both) was less during the first 24 weeks of Rebif® treatment than during the first 24 weeks of Avonex® treatment.

A method was followed by the Central MRI reading facility to avoid double counting of lesions. Images were reviewed for T1 lesions then for T2 lesions and finally reviewed to determine if the T1 and T2 lesions were linked on that scan or an earlier scan. Unique lesions were then counted as lesions that were either T1 active or T2 active or both, but were not double-counted. The mean number of CU active lesions per subjects per scan during the first 24 weeks was calculated as the subject's total number of CU active lesions during the first 24 weeks divided by the subject's total number of scans during the first number of scans during the first 24 weeks.

Three hundred twenty-five subjects in each group underwent repeated MRI scans. Baseline MRI activity was similar between the two treatment groups as was shown in Table 8.

Although the number of CU MRI active lesions present during the study was low overall, subjects treated with Rebif® had significantly fewer CU active lesions during the 24 weeks of treatment compared to those treated with Avonex®, as shown in Table 16 (adjusted mean of 0.8 vs. 1.2, $p<0.001$; median of 0.2 vs. 0.3). The values represent the group adjusted mean (or median) of individual subject's mean lesion numbers per scan. Rebif® treatment resulted in a relative reduction of 33% fewer CU active lesions compared to Avonex® treatment.

Table 16: Mean Number of CU Active Lesions per Subject per Scan

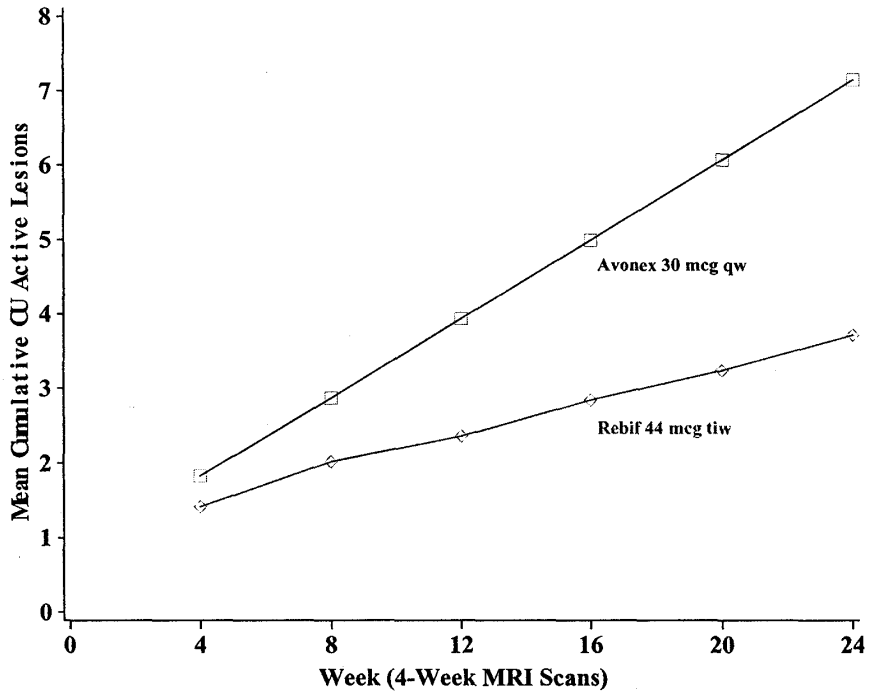
	Rebif	Avonex
N	325	325
Mean (SD)	0.7 (1.7)	1.3 (2.8)
Median	0.2	0.3
Range	0.0, 16.3	0.0, 19.8
Inferential Statistics		
Mean (SEM)	0.8 (0.1)	1.2 (0.1)
	Treatment Comparisons	
Mean Difference (SEM)	-0.5 (0.1)	
95% CI	-0.7, -0.2	
p-value	<0.001	

The mean difference and 95% CI were obtained using a parametric ANCOVA model on raw data with effects for treatment and center with the baseline number of CU active lesions as

the single covariate in the model. The p-value was obtained using a nonparametric ANCOVA model with effects for treatment and center with the baseline number of CU active lesions as the single covariate in the model. These analyses were performed according to the methods prospectively identified in the statistical analytic plan.

Figure 2 presents the cumulative adjusted mean number of CU active lesions by treatment group based on monthly MRI scans. This figure shows a progressive increase in the difference between treatment groups up to the last scan performed at Week 24.

Figure 2: Mean CU Lesions over Time



Exacerbation Rate per Subject

The primary outcome measure only takes into consideration the first clinical exacerbation or relapse. To further examine treatment effect, an assessment of total relapse (exacerbation) rate was performed. Relatively few subjects experienced more than one relapse in the 24 weeks of treatment; 15 on Rebif® and 11 on Avonex®. Overall, subjects treated with Rebif® had a lower exacerbation rate than subjects treated with Avonex® during the 24-week treatment period ($p = 0.22$). The estimated exacerbation rate was 0.293 exacerbations for subjects treated with Rebif® and 0.396 exacerbations for subjects treated with Avonex® during 24 weeks, representing a 26% relative reduction in exacerbations for Rebif® compared to Avonex®.

Table 17: Total Exacerbation Rate per Subject in 24 Weeks

	Rebif	Avonex	
N	339	337	p-value*
Mean (SD)	0.3 (0.5)	0.4 (0.6)	
Median	0	0	
Range	0.0, 2.0	0.0, 2.0	
Exacerbation Rate *	0.293	0.396	0.022

*from a Poisson Regression model with effects for treatment and center

***Reviewer's Note**: Sixty-eight subjects randomized to Rebif® and 110 randomized to Avonex® had one exacerbation, and 15 subjects randomized to Rebif® and 11 randomized to Avonex® had 2 exacerbations. No subject had more than 2 exacerbations during the 24-week period of the study.*

Mean Number of T2 Active Lesions per Subject per Scan

T2 lesions are thought to possibly reflect permanent residual changes to the CNS following an initial inflammatory episode. Table 18 shows that subjects treated with Rebif® had 50% fewer T2 active lesions compared to those treated with Avonex® during the 24-week treatment period.

Table 18: Mean Number of T2 Active Lesions per Subject per MRI Scan

	Rebif	Avonex
N	325	325
Mean	0.4 (1.0)	0.6 (1.2)
Median	0.0	0.2
Range	0.0, 8.5	0.0, 10.2
Mean (SEM)*	0.3 (0.1)	0.6 (0.1)
	Treatment Comparison	
Mean Difference (SEM)*	-0.2 (0.1)	
95% CI	-0.4,-0.1	
p-value**	<0.001	

*estimated using a parametric ANCOVA model on raw data with effects for treatment and center with the baseline number of T2 active lesions as the covariate
**from a nonparametric ANCOVA model with effects for treatment and center with the baseline number of T2 active lesions as the single covariate

Tertiary Endpoints

T1 Gd-positive MRI lesions are viewed as evidence of active blood brain barrier disruption with inflammation that generally persist for 2-8 weeks and may or may not be associated with new T2 lesions. Subjects treated with Rebif® experienced significantly fewer T1 active lesions compared to those treated with Avonex® during the 24-week treatment period (adjusted mean of 0.6 vs. 1.0, median 0.0 vs. 0.2; p <0.001). Rebif® treatment resulted in a relative reduction of T1 active MRI lesions of 40% compared to Avonex® treatment as shown in Table 19.

Table 19: Mean Number of T1 Active Lesions per Subject during 24-Week Treatment

	Rebif	Avonex
N	325	325
Mean (SD)	0.6 (1.5)	1.1 (2.6)
Median	0.0	0.2
Range	0.0, 17.7	0.0, 18.3
	Treatment Comparison	
Mean Difference (SEM)*	-0.4 (0.1)	
95% CI*	-0.6, -0.2	
p-value**	<0.001	

*estimated using a parametric ANCOVA model with effects for treatment and center with the baseline number of T1 active lesions as the covariate
**from a non-parametric ANCOVA model with effects for treatment and center with the baseline number of T1 active lesions as the single covariate

Subjects treated with Rebif® also had a significantly smaller proportion of scans with active CU lesions compared to those treated with Avonex®. Rebif® treatment resulted in a 36% relative reduction in mean, and 50% relative reduction in median CU active MRI scans per subject relative to Avonex® treatment. This proportion of CU active scans is derived from the proportion of T2 active scans per subject (adjusted mean of 14.8% for Rebif® vs. 26.8% for Avonex®, median 0.0% for Rebif® vs. 16.7% for Avonex®; p<0.001) for a relative reduction of 45% in mean T2 active scans per subject for Rebif® vs. Avonex® and from the proportion T1 active scans per subject (adjusted mean of 20.9% for Rebif® vs. 33.2% for Avonex®, median 0.0% for Rebif® vs. 20% for Avonex®; p<0.001) for a 37% relative reduction in mean T1 active scans per subject for Rebif® relative to Avonex® treatment.

Table 20: Percentage of CU Active Scans per Subject During 24-Week Treatment

	Rebif	Avonex
N	325	325
Mean	23.6 (30.5)	38.2 (36.4)
Median	16.7	33.3
Range	0.0, 100.0	0.0, 100.0
Mean (SEM)*	24.0 (1.7)	37.3 (1.6)
	Treatment Comparison	
Mean Difference (SEM)*	-13.3(2.3)	
95% CI	-17.8, -8.8	
p-value**	<0.001	

*estimated using a parametric ANCOVA model with effects for treatment and center with the baseline number of CU active lesions as the covariate
**from a non-parametric ANCOVA model with effects for treatment and center with the baseline number of CU active lesions as the single covariate

Rebif® treatment resulted in a relative increase of 45% in subjects with no CU active lesions compared to Avonex®. The percentage of Rebif®-treated subjects with no active CU lesions during the 24 weeks on study was slightly greater than the percentage of subjects with no active CU lesions during the four weeks prior to treatment (48% vs. 45%) despite the 6-fold increase in observation time and number of scans performed. The percentage of subjects on Avonex® with CU lesion activity during the 24 weeks on study continued to increase such that the percentage of subjects with no CU active lesions was 33% after 24 weeks of treatment compared to the baseline percentage of 45%. This was composed of data regarding absence of both T2 active lesions as well as T1 active lesions, shown in Table 21.

Table 21: Subjects With Inactive and Active MRI Scans During 24-Week Treatment Period

	Rebif	Avonex
	N = 325	N = 325
	N (%)	N (%)
No CU Active Lesions	157 (48.3)	108 (33.2)
CU Active Lesions	168 (51.7)	217 (66.8)
	Treatment Comparison	
Odds Ratio*	2.0	
95% CI*	1.4, 2.8	
p-value*	<0.001	
No T2 Active Lesions	196 (60.3)	141 (43.4)
T2 Active Lesions	129 (39.7)	184 (56.6)
	Treatment Comparison	
Odds Ratio*	2.1	
95% CI*	1.5, 3.0	
p-value*	<0.001	
No T1 Active Lesions	177 (54.5)	123 (37.8)
T1 Active Lesions	148 (45.5)	202 (62.2)
	Treatment Comparison	
Odds Ratio*	2.1	
95% CI*	1.5, 3.0	
p-value*	<0.001	

*from a logistic regression model with effects for treatment and center

One hundred ninety-six subjects treated with Rebif® had no T2 active lesions compared to 141 subjects treated with Avonex® during the 24 week treatment period. The percentage of Rebif® treated subjects with no active T2 lesions during the 24 weeks on study was similar to the percentage with no active T2 lesions during the 4 weeks prior to treatment (60% vs. 62%), whereas the percentage of Avonex® treated subjects with no active T2 lesions during the 24 weeks on study was less than the percentage with no active T2 lesions during the 4 weeks prior to treatment (43% vs. 63%). One hundred seventy-seven subjects treated with Rebif® had no T1 active lesions compared to 123 subjects treated with Avonex® during the 24 week treatment period. The percentage of Rebif® treated subjects with no active T1 lesions during the 24 weeks on study was similar to the percentage of subjects with no active T1 lesions during the 4 weeks prior to treatment (55% vs. 57%), whereas the percentage of Avonex® treated subjects with no active T1 lesions during the 24 weeks on study was less than the percentage of subjects with no active T1 lesions during the 4 weeks prior to treatment (38% vs. 55%).

Exploratory Analyses

Rebif® prolonged the time to the first clinical exacerbation during the 24-week treatment period compared to Avonex® (p =0.001; hazard ratio 0.63). The Kaplan Meier estimate of

the 20th percentile time to first relapse was 3.9 months for Rebif® treated subjects and 2.9 months for Avonex® treated subjects.

Exacerbation severity was assessed by changes in the EDSS and KFS disability scores, and the Activities of Daily Living (ADL) scale score. Approximately two-thirds of all relapses were graded as moderate or severe, i.e. ≥ 1 EDSS point or ≥ 2 points on the KFS scale. There was no difference in the proportions of subjects in either group in terms of relapse severity, although the absolute number of relapses in each category was less in the Rebif® group than in the Avonex® group as shown in Table 22.

Table 22: Exacerbations by Severity

	Rebif	Avonex
Number of Exacerbations (%)	98	132
Severity by EDSS/KFS		
Mild	27 (27.6)	40 (30.3)
Moderate	39 (39.8)	49 (37.1)
Severe	23 (23.5)	30 (22.7)
Not available	9 (9.2)	13 (9.8)

The rate of steroid use for MS exacerbations was 0.094 courses per subject during the 24 weeks in the Rebif® group and 0.177 courses per subject in the Avonex® group (p=0.004). Overall, approximately one-third of relapses in the Rebif® subjects were treated with steroids for their relapses, whereas approximately one-half of relapses in the Avonex® subjects were treated with steroids.

Reviewer's Note: *These data suggest that the physicians unblinded to treatment (who decided what the appropriate treatment for an exacerbation should be) were biased toward treating exacerbations that occurred in the Avonex® treatment arm more often than exacerbations occurring in the Rebif® treatment arm. However, these data also suggest that the blinded evaluators were not biased in the way they rated exacerbations between the two treatment groups, i.e., they did not rate those that occurred in subjects treated with Rebif® as less severe and the ones occurring in subjects treated with Avonex® as more severe.*

Additional Exploratory Analyses Performed

Change in EDSS Score from Baseline to 24 Weeks:

An exploratory analysis was performed on change in EDSS score from baseline to 24 weeks. The Wilcoxon rank sum test yielded a statistically significant p-value of 0.041, favoring Rebif®. Any subjects who were experiencing an exacerbation at six months at the time of the neurologic evaluation would contaminate this analysis group. It will be instructive to re-do this analysis when the 9 month data are available.

Subjects Who Experienced a Clinical Exacerbation Within the First 3 Months:
There were 120 subjects in this category, 53 in the Rebif® group (15.6%) and 67 in the Avonex® group (19.8%). The distributions of time to first exacerbation were similar for the two treatment groups. For the Rebif® group the median was 48 days with a range from 1 to 89 days; for the Avonex® group the median was 48 days with a range from 2 to 89 days.

Safety Analyses

Serious Adverse Events and Deaths

No deaths occurred during the 24-week study period.

Twenty-four serious adverse events occurred in 22 subjects (3.3%); 14 in the Rebif® group and 10 in the Avonex® group. In each of the two treatment groups, three of the serious adverse events were deemed by the investigators as at least possibly related to the study treatment. The serious adverse events are shown in Table 23.

Table 23: Serious Adverse Events

Body System Preferred Term	Rebif N = 339 No. of Subjects (%)	Avonex N = 337 No. of Subjects (%)
Total	13 (3.8%)	9 (2.7)
Body as a Whole	4 (1.2)	3 (0.9)
Death Fetal	2 (0.6)	1 (0.3)
Allergic Reaction	1 (0.3)	1 (0.3)
Chest Pain	1 (0.3)	1 (0.3)
Syncope	0	1 (0.3)
Psychiatric Disorders	3 (0.9)	2 (0.6)
Depression	0	2 (0.6)
Depresssion Aggravated	1 (0.3)	0
Emotional Lability	1 (0.3)	0
Suicide Attempt	1 (0.3)	0
GI Disorders	2 (0.6)	1 (0.3)
Diarrhea	0	1 (0.3)0
Enteritis	1 (0.3)	0
Esophagitis	1 (0.3)	0
Resistance Mechanism Disorders	2 (0.6)	0
Abscess	1 (0.3)	0
Otitis Media	1 (0.3)	0
CV Disorders	0	1 (0.3)
ECG Abnormal	0	1 (0.3)
CNS and PNS Disorders	0	1 (0.3)
MS Aggravated	0	1 (0.3)
Neoplasm	1 (0.3)	0
Breast Neoplasm (malignant)	1 (0.3)	0
Respiratory Disorders	0	1 (0.3)
Epiglotitis	0	1 (0.3)
Secondary Terms	1 (0.3)	0
Fall	1 (0.3)	0
White Cell and Res. Disorders	1 (0.3)	0
Lymphopenia	1 (0.3)	0

Reviewer's Note: A *review of the narratives for the Serious Adverse Events presented in Table 23 reveals the following:*
For subjects randomized to Rebif ®,

- *Subject XXXXXXXXXX was hospitalized for an allergic reaction with severe edema of the face and neck beginning approximately 3 months after starting Rebif®. She required treatment with epinephrine and steroids, including IM methylprednisolone. No etiology was found. The investigator stated that the event was unrelated to Rebif® and the subject was continued on Rebif®.*
- *Subject XXXXXXXXXX was hospitalized for what was thought to be a severe allergic reaction to solumedrol about 2 months after beginning Rebif®. The investigator considered this event unlikely related to Rebif®. Rebif® was continued.*
- *Subjects XXXXXXXXXX and XXXXXXXXXX experienced spontaneous abortions while taking Rebif®. In one case the investigator felt the miscarriage was unrelated to Rebif® (Rebif® dose was interrupted in subject XXXXXXXXXX); in the other case the investigator attributed it as possibly related to Rebif® and Rebif® was discontinued (subject XXXXXXXXXX).*
- *Subject XXXXXXXXXX was hospitalized for depression with suicidal ideation. The investigator thought the depression was possibly related to Rebif® but continued the Rebif®.*
- *Subject XXXXXXXXXX was hospitalized twice for emotional distress which the investigator thought was unlikely related to Rebif® and continued the Rebif®.*
- *Subject XXXXXXXXXX was hospitalized for a suicide attempt about 3 weeks after beginning Rebif®. The investigator felt the depression and suicide attempt were unrelated to Rebif®. Rebif® was continued, and treatment with Zoloft begun.*
- *Subject XXXXXXXXXX developed a Grade 3 lymphopenia approximately 5.5 months after starting Rebif®. The Rebif® was discontinued, and her lymphocyte count returned to normal.*

For subjects randomized to Avonex®:

- *Subject XXXXXXXXXX had a spontaneous abortion after approximately 3 months on Avonex® that the investigator considered unlikely to be related to Avonex® treatment.*
- *Subject XXXXXXXXXX was a 39 year old woman who experienced palpitations, premature atrial contractions and ST-T wave changes on ECG that the investigator considered unrelated to Avonex®.*
- *Subject XXXXXXXXXX was hospitalized for a severe depressive episode approximately 2 months after starting Avonex®. The investigator considered this event as probably related to Avonex®, but the drug was apparently continued.*
- *Subject XXXXXXXXXX was hospitalized for an increased severity of her depression. It began to worsen about 2 months after beginning Avonex®. The investigator thought the increased depression was probably related to Avonex®, and her Avonex® dose was reduced. She was begun on the antidepressant Celexa, and was also reported to be taking Klonopin and the antipsychotic Seroquel.*

Reviewer's Note: *These serious adverse event narratives confirm FDA's concerns that investigators may not be appropriately aware of some of the potential risks of interferon treatment, including the abortifacient effects of the interferons, the fact that treatment with*

interferons may cause or exacerbate depression sometimes to a degree where the depression becomes life-threatening, that there be rare cardiac adverse events associated with interferon administration, and that there may be rare severe allergic reactions associated with the administration of Rebif®.

Important Rare Serious Adverse Events

There have been several rare, but important, serious adverse events observed following treatment with Rebif®, some of which were life-threatening that have not to date been reported to FDA following treatment with either of the other interferon-betas approved for the treatment of MS, Avonex® or Betaseron®. These events consist of two occurrences of anaphylaxis, one occurrence of fulminant hepatic failure requiring liver transplantation, one occurrence of a Stevens-Johnson syndrome, one occurrence of what was deemed to be a life-threatening cardiac arrhythmia, and two occurrences of erythema multiforme. These serious adverse events did not occur in the study that is the focus of this review. The severe allergic reaction described in the serious adverse event narratives for Study XXXXXXXXXX indicates that the subject developed severe edema of the face and neck of unknown etiology, and that she was maintained on treatment with Rebif®.

One occurrence of anaphylaxis thought to be due to Rebif® occurred and was reported in study GF 6789 as described in the "Overview of Prior Clinical Studies of Rebif®." The other serious adverse events occurred in subjects being treated with marketed drug, and thus were filed as post-marketing safety reports. One case of anaphylaxis temporally related to Rebif® administration was initially reported as a letter to the Editor of the journal Neurology.

A report of fulminant hepatic failure was submitted to FDA in September 2000 as a post-marketing serious adverse event report from a physician in Canada initially reported to the Applicant. A 59-year-old woman with RRMS was begun on Rebif® 11 µg 3 x per week in July 2000, 1.25 years after the diagnosis of MS was made. Rebif® was stopped for 6 days at the end of July because of "minor side effects." When she was evaluated in the clinic on August 22, 2000, she complained of increasing fatigue, nausea, flatus and insomnia. An abdominal ultrasound was said to show gallstones and possible blockage of the biliary duct. On August 25th her blood work was reported to show normal hematology and "extremely elevated transaminases and bilirubin" that were > 10 times normal. She had negative serology for hepatitis A, B and C. She was admitted to a hospital on September 3rd and was transferred to a tertiary care facility on September 7th in hepatic failure. She underwent a liver transplant on September 10, 2000. The reporting physician felt that the event probably represented an autoimmune reaction due to Rebif®, and the liver histopathology supported a diagnosis of hepatic necrosis due to an autoimmune or toxic drug reaction. However, a correction published in the journal Neurology on December 11, 2001 stated "…Yoshida et al. reported a patient who developed fulminant liver failure requiring urgent liver transplantation, 7 weeks after commencing interferon ß-1a (Rebif®) …the authors have received new information that she had been started on nefazodone in February 2000. Nefazodone has recently been reported in association with acute liver failure…Interferon ß1-a appears to be temporally implicated in the patient's liver failure but the authors cannot exclude the possibility that nefazodone may have been a factor or cofactor."

Another serious adverse event report was that of a 45 year old woman who had been treated with Rebif® 22 µg 2 x per week for one year, who developed a cardiac arrhythmia, considered life-threatening by the reporting physician, and for which Rebif® treatment was discontinued. Myocardial infarction and myocarditis were also suspected, but the reporting physician provided no specific information regarding these diagnoses.

The report of a Stevens-Johnson syndrome occurred in a 54-year-old woman with no known risk factors and who was not taking any concomitant medications. Her severe rash developed shortly after beginning Rebif®, although the exact number of doses was not specified in the expedited report submitted to FDA. The Applicant reports no other cases of Stevens-Johnson syndrome in their database. Two cases of erythema multiforme have been reported, however, and have been assessed by the reporting physicians as being "possibly" related to Rebif® treatment. Note that classical erythema multiforme has non-drug etiologies, and must be differentiated from Stevens-Johnson syndrome, which is usually drug-related.

Severe Adverse Events

There were 69 adverse events rated as severe in 44 subjects in the Rebif® treatment group, and two rated as "life threatening" in two subjects. There were 71 severe adverse events in 52 subjects in the Avonex® treatment group, and one rated as "life-threatening."

The "life-threatening" adverse events in the Rebif® arm were both psychiatric disorders: one was "depression aggravated" and one was a "suicide attempt." The adverse event in the Avonex® group that was considered to be life-threatening was an allergic reaction to gadolinium. Other notable severe adverse events are shown in Table 24. They were selected because of concerns from the body of evidence that have arisen on the use of the ß-interferons, specifically related to generalized and local injection site reactions, psychiatric disturbances (particularly depression), hepatic dysfunction, cytopenias (particularly of WBCs) and thyroid disorders.

Table 24: Selected Severe Adverse Events

Preferred Term	Rebif N = 339	Avonex N = 338
Influenza-Like Symptoms	2 (0.6)	11 (3.3)
Headache	7 (2.1)	12 (3.6)
Fatigue	4 (1.2)	2 (0.6)
Fever	0	1 (0.3)
Rigors	0	1 (0.3)
Injection Site Pain	2 (0.6)	0
Depression	2 (0.6)	4 (1.2)
Emotional Lability	2 (0.6)	0
Depression Aggravated	1 (0.3)	0
Myalgia	2 (0.6)	2 (0.6)
Alopecia	1 (0.3)	0
SGPT increased	2 (0.6)	0
SGOT increased	0	0
Hepatic Enzymes Increased	1 (0.3)	3 (0.9)
Hepatitis	0	0
Gamma-GT Increased	0	0
Hepatocellular Damage	0	0
Creatine Phosphokinase Inc.	2 (0.6)	0
Leukopenia	0	0
Lymphopenia	2 (0.6)	1 (0.3)
Granulocytopenia	0	0
Thyroid Disorder	1 (0.3)	0

Many of the above events were much more frequently reported and graded as “mild” or “moderate” in severity. Selected adverse events that are known to be associated with interferon-β administration are shown in Table 26.

Analysis by Body System and Event

Review of the overall adverse event profile for Rebif® included in this submission revealed it to be similar to that observed with the β-interferons to date. It was also generally similar to the adverse events and their frequencies as reported in the current package inserts for Avonex® and Betaseron®, with only a few exceptions, discussed elsewhere. Adverse events that occurred in ≥ 5% of subjects in either the Rebif® or Avonex® treatment group in Study XXXXXXXXXX are shown in Table 25.

Table 25: Adverse Events Reported in ³5% of Subjects

Total with Adverse Events	331 (97.6%)	321 (95.3%)
Body System		
Preferred Term		
Body as a Whole	255 (75.2)	268 (79.5)
Influenza-Like Symptoms	141 (41.6)	164 (48.7)
Headache	114 (33.6)	101 (30.0)
Fatigue	53 (15.6)	55 (16.3)
Back Pain	25 (7.4)	29 (8.6)
Fever	15 (4.4)	23 (6.8)
Abdominal Pain	20 (5.9)	11 (3.3)
Rigors	10 (2.9)	21 (6.2)
Application Site Disorders	273 (80.5)	82 (24.3)
Injection Site Inflammation	146 (43.1)	15 (4.5)
Injection Site Reaction	111 (32.7)	31 (9.2)
Injection Site Pain	62 (18.3)	31 (9.2)
Injection Site Bruising	26 (7.7)	12 (3.6)
Resistance Mechanism Disorders	154 (45.4)	167 (49.6)
Rhinitis	58 (17.1)	52 (15.4)
Upper Resp. Tract Infection	34 (10.0)	44 (13.1)
Sinusitis	31 (9.1)	24 (7.1)
Infection, Viral	20 (5.9)	33 (9.8)
Urinary Tract Infection	14 (4.1)	21 (6.2)
Pharyngitis	14 (4.1)	20 (5.9)
Psychiatric Disorders	96 (28.3)	86 (25.5.)
Depression	38 (11.2)	45 (13.4)
Insomnia	44 (13.0)	35 (10.4)
CNS and PNS Disorders	89 (26.3)	82 (24.3)
Dizziness	25 (7.4)	23 (6.8)
Hypertonia	11 (3.2)	18 (5.3)
Gastrointestinal Disorders	81 (23.9)	89 (26.4)
Nausea	30 (8.8)	25 (7.4)
Musculoskeltal System Disoders	61 (18)	56 (16.6)
Myalgia	35 (10.3)	39 (11.6)
Arthralgia	21 (6.2)	18 (5.3)
Liver and Biliary System Disorders	47 (13.9)	22 (6.5)
SGPT Increased	26 (7.7)	9 (2.7)
SGOT Increased	21 (6.2)	3 (0.9)

There were no increases in infections in either treatment group (35% overall infection rate during 6 months in both treatment groups) associated with decreases in white blood cells. There were more severe influenza-like symptoms in the Avonex® group, although overall, the incidence was quite similar. Depression was also more frequent in the Avonex® group, although other psychiatric disturbances classified as "emotional lability" were somewhat more frequent in the Rebif® group.

Abnormalities of liver function tests and decreases in white blood cell counts and injection site reactions, including pain, were more common in the Rebif® group, although most were mild to moderate in severity.

Table 26: Selected Adverse Events by Severity

	Rebif N = 339			Avonex N = 338		
	Mild	**Moderate**	**Severe**	**Mild**	**Moderate**	**Severe**
Preferred Term	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)
Influenza-Like Symptoms	98 (29)	45 (13)	2 (0.6)	109 (32)	53 (16)	11 (3.3)
Injection Site Pain	45 (13)	15 (4)	2 (.6)	30 (8.9)	1 (0.3)	0
Depression	11 (3.2)	25 (7.4)	2 (0.6)	23 (6.8)	18 (5.3)	4 (1.2)
Insomnia	29 (8.6)	14 (4.1)	1(0.3)	24 (7.1)	10 (3.0)	1 (0.3)
Anxiety	3 (0.9)	7 (2.1)	0	3 (0.9)	7 (2.1)	1 (0.3)
Emotional Lability	2 (0.6)	5 (1.5)	2 (0.6)	1 (0.3)	3 (0.9)	0
Depression Aggravated	0	0	1 (0.3)	0	0	0
Suicide Attempt	0	0	0	0	0	0
SGPT Increased	12 (3.5)	12 (3.5)	2 (0.6)	4 (1.2)	5 (1.5)	0
SGOT Increased	11 (3.2)	10 (2.9)	0	2 (0.6)	1 (0.3)	0
Hepatic Enzymes Increased	9 (2.7)	6 (1.8)	1 (0.3)	3 (0.9)	1 (0.3)	3 (0.9)
Hapatocellular Damage	0	1 (0.3)	0	0	0	0
Leukopenia	6 (1.8)	5 (1.5)	0	1 (0.3)	0	0
Lymphopenia	2 (0.6)	5 (1.5)	2 (0.6)	0	0	1 (0.3)
Granulocytopenia	8 (2.4)	0	0	1 (0.3)	0	0
Thyroid Disorder	3 (0.9)	0	1 (0.3)	3 (0.9)	0	0

Pregnancies

Five pregnancies occurred during this study, four in the Rebif® treatment arm, and one in the Avonex® treatment arm. Three of the pregnancies (2 in the Rebif® group, 1 in the Avonex® group) ended in spontaneous abortions; one in the Rebif® group was terminated by a therapeutic abortion, and one pregnancy was carried to term with birth of a healthy, full-term infant.

Development of Antibodies to Interferon-ß

The results of these assays were not included in this submission because the Applicant stated prospectively that the assays would not be performed until all Week 48 samples were obtained. The assays have not been performed as of the end of November 2001.

Assessment and Conclusions

- This BLA supplement consists of a single randomized, unblinded, active treatment, comparative multicenter study conducted in 677 subjects with relapsing-remitting MS that utilized blinded evaluators for both the neurologic examinations and for interpretation of the MRI findings. The study was designed to compare the efficacy and safety of 44 µg of Rebif® administered SC 3 x per week *vs.* 30 µg Avonex® administered IM once weekly in delaying or preventing the occurrence of clinical exacerbations in subjects who had experienced at least two clinical exacerbations during the previous two years.

- The BLA supplement is comprised of complete detailed safety data from Study XXXXXXXXXX (the comparative study) and of limited selected safety data from 39 studies that include 4469 subjects with MS who have been treated with Rebif®. Specifically, the supplement contains the safety data from the 677 subjects enrolled in the comparative study as well as selected safety data from two placebo-controlled studies of Rebif® treatment for relapsing-remitting MS (560 subjects in the PRISMS study [445 subjects who remained in the study for four years] that compared 22 µg and 44 µg of Rebif® administered 3 x per week vs. placebo for 2 years, and 293 subjects in the OWIMS study that compared 22 µg and 44 µg of Rebif® administered once per week vs. placebo for 48 weeks).

- The primary study endpoint was the proportion of subjects who were exacerbation-free following 24 weeks of treatment.

- Three secondary endpoints, two based on MRI parameters and one based on clinical symptoms, identified prospectively by the Applicant were evaluated, along with three tertiary endpoints, all based on MRI parameters.

- Demographics and baseline neurologic evaluations were well-balanced between the treatment groups.

- The primary endpoint, proportion of subjects who were exacerbation-free following 24 weeks of treatment, demonstrated Rebif® 44 µg administered SC 3 x per week to be superior to Avonex® 30 µg administered IM 1 x per week ($p<0.001$, relative risk of 1.3, with a 95% confidence interval of 1.1, 1.6). At the time of the analyses conducted following 24 weeks of treatment, 254 of 339 subjects (74.9%) in the Rebif® treatment group were exacerbation-free, compared with 214 of 338 subjects (63.3%) in the Avonex® treatment group. CBER-conducted analyses on the primary endpoint confirmed the analyses done by the Applicant.

- The three secondary endpoints prospectively identified by the Applicant and ranked in order of importance all showed statistically significant benefits of Rebif® compared to Avonex®, and included the mean number of combined unique (CU) T1 + T2 active MRI lesions per subject per scan ($p<0.001$), the total exacerbation count per subject ($p=0.022$) and the mean number of T2 active lesions per subject per MRI scan ($p<0.001$). The mean

number of combined unique MRI lesions showed a progressive difference between the treatment groups up to the last scan performed at 24 weeks. The three tertiary endpoints also showed statistically significant benefit of Rebif® compared to Avonex® treatment on these MRI parameters ($p<0.001$).

- The observed safety profile for Rebif® was similar to the safety profile observed for Avonex®, with the exceptions of increased frequency of liver function test abnormalities, decreases in white blood cell counts and injection site reactions that were generally mild to moderate in severity. These adverse events have been observed to occur at similar rates in other studies of Rebif® administered SC and are common to all the interferon-betas.

- Relatively small numbers of subjects discontinued from both treatment groups due to adverse events – 11 (of 339) in the Rebif® group and 3 (of 338) in the Avonex® group.

- Study conduct overall appeared to have been done in compliance with Good Clinical Practices. An FDA-conducted inspection of three clinical study sites revealed no significant deficiencies in the conduct of the study. The Applicant was informed by the principal investigator at site #238 of some allegedly fraudulent recording of data. The Applicant performed a site audit, had an independent audit of the site conducted, and based on their findings recommended that the data from site #238 be excluded from the efficacy analyses. FDA concluded the Applicant and principal investigator had taken all necessary measures and that no additional action by FDA was warranted. Exclusion of the data from this study site had no impact on the robustness of the overall study conclusions.

Conclusions:

Based on the results of this randomized, multicenter study utilizing an active comparator control, Rebif® administered at a dose of 44 µg 3 x per week appears to have superior clinical efficacy to Avonex® administered at the indicated dose of 30 µg IM 1 x per week for increasing the proportion of subjects with relapsing-remitting multiple sclerosis who will be exacerbation-free following 6 months of treatment. The clinical effect is moderate, while the level of statistical significance is very robust and is supported by MRI effects.

The safety profile of Rebif® is similar to that of the other interferon betas, with the exception of its association with more frequent abnormalities of liver function tests, cytopenias and injection site reactions, particularly when compared with the frequencies reported in the Avonex® label.

This study should be viewed as confirming the efficacy of Rebif® in reducing the frequency of clinical exacerbations in subjects with relapsing-remitting MS that was previously demonstrated in the original BLA submitted in 1998. The results of the 48-week data from this study will aid in determining whether the superior clinical efficacy of Rebif® compared to Avonex® demonstrated in this study is sustained beyond 6 months.

Financial Disclosure Statements

FDA Forms 3454 were submitted for 55 of the 56 principal investigators who participated in study XXXXXXXXXX certifying their absence of financial interests as defined in 21CFR54.2(a), (b) and (f).

One principal investigator and four sub investigators who participated in study XXXXXXXXXX disclosed certain financial arrangements with the Applicant, none of which enrolled a large enough proportion of the study population to affect the study results.

XXXXXXXXXX was a sub investigator and evaluating physician for Dr. Yves Lapierre at site 191 (Montreal Neurological Institute, Montreal, Quebec, Canada) at which 16 subjects were enrolled. Serono, Inc. contracted with XXXXXXXXXX to perform atrophy measures and analyses on a subset of MRI data generated at the XXXXXXXXXX MS/MRI Research Centre from Serono protocol XXXXXXXXXX (the PRISMS study). XXXXXXXXXX. The subject enrollment at this site represented only 2.4% of the study population.

Dr. Reinhard Hohlfeld was the principal investigator and XXXXXXXXXX was a sub investigator and treating physician at site 271 (Klinikum Grosshadern der Ludwig-Maximilians-Universität, Munchen, Germany). Dr. Hohlfeld's site enrolled 9 subjects, representing 1.3% of the study population. XXXXXXXXXX.

XXXXXXXXXX was a co-investigator with XXXXXXXXXX and was the treating physician at site 297 (MS Clinical Research Group, XXXXXXXXXX, Vancouver, BC, Canada), at which 12 subjects were enrolled, representing 1.8% of the study population. XXXXXXXXXX.

XXXXXXXXXX was a sub investigator for Dr. Bever at site 222 (University of Maryland Hospital, Baltimore, MD) and back up evaluating physician who was involved in one subject assessment. Dr. Bever's site enrolled 16 subjects, representing 2.4% of the study population. XXXXXXXXXX. Samples from the 16 subjects enrolled at his site will be analyzed by quantitative PCR.

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